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P1. REPLICABLE SUBGROUPS OF YOUTH WITH DISTRESSING PSYCHOTIC-LIKE EXPERIENCES VIA MULTIMODAL DATA FUSION

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Background: The mental health of young people is a growing public health concern, yet interventions in schools, universities, and health systems have shown limited effectiveness. A key challenge is identifying youth most vulnerable to adverse neurodevelopmental, cognitive, and psychiatric outcomes. Psychotic-like experiences (PLEs) have emerged as promising early risk markers. While relatively common in late childhood and adolescence (present in ~20% of youth), PLEs are far more prevalent (~50%) among youth with existing mental health conditions. Distressing PLEs are associated with a 57–92% likelihood of later psychiatric illness and with suicidal thoughts and behaviors. Youth reporting PLEs also show lower cognitive performance, greater environmental adversity, and atypical brain development. However, the relative contributions of these factors to long-term outcomes remain unclear. This study aimed to identify data-driven multimodal subgroups of youth experiencing distressing PLEs and to examine how the features defining these subgroups relate to the presence of PLEs both cross-sectionally and longitudinally.

Methods: Using data from the Adolescent Brain Cognitive Development Study, we examined 1716 youth reporting distressing PLEs (50.4% female) alongside a normative group without PLEs ($n = 5225$; 50.9% female). Youth with PLEs were divided into independent discovery and replication sets ($n = 858$ each; 49.7% female in discovery, 51.3% in replication). Cognitive, environmental, and structural and functional MRI were integrated using Similarity Network Fusion (SNF) to identify data-

driven subgroups. Cluster validity was assessed through resampling, stability indices, and replication. Subgroup differences were tested on out-of-model measures, including psychopathology (assessed via Child Behavior Checklist) and white matter integrity. Longitudinal analyses examined PLE persistence across three time points (baseline, 1-year, 2-year) using latent growth modeling. Cognitive performance at baseline and 2-year follow-up was evaluated to determine whether subgroup differences persisted over time.

Results: Multimodal integration revealed five distinct and reproducible subgroups of youth in distress. Subgroups were highly consistent across discovery and replication samples, with approximately 90% overlap among the top 15% of contributing features. Significant differences were observed across cognitive, environmental, and neuroimaging features ($pFDR < 0.05$), with the largest effects in cognitive performance, environmental adversity, cortical thickness, and subcortical volume. These subgroup distinctions extended to out-of-model measures, including psychopathology and white matter integrity ($pFDR < 0.05$), demonstrating both clinical and biological relevance. Cognitive differences at baseline persisted over time (Group $\times$ Time: $F(3,678) = 20.02$, $p < 0.001$). Subgroup membership was associated with distinct longitudinal PLE trajectories ($\chi^2(8, n = 1545) = 33.29$, $p < 0.001$), with persistently high PLEs significantly overrepresented in the most impaired subgroup. This highlights that early differences in cognitive, environmental, and neurobiological profiles predict divergent patterns of symptom persistence.

Conclusions: These findings support multimodal data integration as a powerful approach for capturing heterogeneity among youth at risk for adverse mental health outcomes. Incorporating cognitive, environmental, and neuroimaging features enhances the identification of youth most vulnerable to persistent impairments, including ongoing distressing PLEs and cognitive impairments, and provides insight into the complex interplay of factors that shape risk trajectories. Future research should aim to replicate these findings in independent cohorts and explore how multimodal approaches can inform early detection, personalized

intervention strategies, and preventative programs to mitigate long-term psychiatric risk in youth populations.

Keywords: Adolescent Brain Cognitive Development study, psychotic-like experiences, multimodal data

Disclosure: Nothing to disclose.

P2. WORKING MEMORY AND MOTIVATION IN ADOLESCENTS WITH AND WITHOUT DEPRESSION

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Background: Cognitive and motivational neurobehavioral systems undergo dynamic development during adolescence. At the same time, psychiatric illnesses marked by cognitive and motivational deficits, such as depression, emerge during this developmental period. To better understand the co-occurrence of motivational and cognitive impairments in adolescents with depression (AD), we report a neurobehavioral analysis of working memory and motivated working memory in AD and neurotypical peers (NP).

Methods: 30 AD ($F = 19$, $M = 12$, $NB = 2$) and 34 NP ($F = 22$, $M = 12$) completed a motivated spatial working memory (sWM) task in the fMRI scanner. Participants were instructed to keep spatial locations in mind, and in some trials could win or lose money based on sWM performance. Participants performed non-incentivized (baseline) and incentivized sWM trials. Incentives were cued prior to each trial, or presented in a contextual, non-cued manner at the start of a block of trials. Angular bias and precision of sWM performance were analyzed between groups. Neuroimaging used multi-band sequences that were preprocessed with Human Connectome Project minimal preprocessing pipelines as implemented in QuNex. Whole-brain analyses were conducted using QuNex and Permutation Analysis of Linear Models (PALM).

Results: Examination of sWM memory precision demonstrated a significant main effect of condition ($F = 59.7$, $p < 0.001$), and non-significant group effects and group*condition interaction. Overall, both groups improved sWM performance in response to incentives. Examination of sWM bias demonstrated a significant group*condition interaction ($F = 2.65$, $p < 0.05$), driven by AD relying more on a categorical strategy for sWM maintenance during the cued loss condition, compared to NP. Whole-brain neuroimaging data showed a significant group*sWM effect in the intraparietal sulcus (IPS), anterior parietal cortex and frontal eye fields (FEF) ($p < 0.05$, fwe-protected, 1000 permutations). AD had relatively decreased BOLD signal during sWM encoding and delay in the anterior parietal cortex and FEF, compared to NP. AD had relatively increased BOLD signal during sWM in the posterior IPS, compared to NP.

Conclusions: Adolescents with and without depression demonstrated differences in the use of categorical bias when performing an incentivized spatial working memory task. Early analysis of neuroimaging data suggests that AD and NP differentially engage neural circuits within posterior IPS, anterior parietal cortex and FEF during sWM. Future work can examine neural circuitry of incentivized sWM in AD and NP, as well as links between symptoms, sWM performance and neural engagement, in order to identify possible biomarkers for tracking illness course and treatment development.

Keywords: Adolescent Depression, Cognition, Human Neuroimaging, motivation

Disclosure: Boehringer Ingelheim, Grant, Self, Boehringer Ingelheim, Honoraria, Self.

P3. PREMORBID HYPERPERFUSION IN SUBCORTICAL REWARD CIRCUITS PREDICTS YOUNGER AGE OF FIRST ADOLESCENT SUBSTANCE USE

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Background: Adolescence is a period of development characterized by rapid changes in brain structure, function, and neurochemistry, as well as normative declines in cerebral blood flow (CBF) that reflect increasing neural efficiency. Unlike structural changes, perfusion in cortical regions, including the prefrontal cortex, is steep. In contrast, perfusion in subcortical reward circuitry is more protracted, suggesting sustained, elevated metabolic demand relative to cortical control systems. This developmental “reward-control” imbalance can increase susceptibility to risk-taking and may predispose vulnerable youth to earlier initiation of substance use.

Methods: Multimodal neuroimaging data were acquired using a Siemens 3T TIM Trio and pseudo-continuous arterial spin labeling (PCASL) to quantify CBF at rest. Participants were healthy, substance-naïve adolescents aged 13–14 years ($n = 52$, 48% female) who underwent brain imaging annually for 3 years and were evaluated quarterly for substance use. For the present report, only baseline imaging data were analyzed. During quarterly follow-ups, $n = 25$ participants initiated alcohol or other substance use, while $n = 27$ remained substance-naïve. Differences in global CBF averaged over gray matter and in subcortical regions, covaried for sex, were assessed.

Results: While there were no group differences in global gray matter CBF, adolescents who later initiated alcohol or substance use had significantly higher CBF in subcortical regions, right caudate, thalamus, hippocampus, left pallidum, and bilateral putamen and ventral diencephalon compared with non-initiators (all $p < 0.05$, corrected). Within the initiator group, higher bilateral pallidal perfusion was negatively associated with a younger age of first use ($p < 0.05$). Initiator group differences in subcortical perfusion were observed, even in the absence of differences in risk-taking, impulsivity, or clinical measures.

Conclusions: Adolescents who later initiated substance use exhibited higher baseline perfusion in reward processing circuitry biased toward habit formation (e.g., putamen, pallidum), rather than classic limbic reward regions (e.g., nucleus accumbens, amygdala). These findings suggest evidence for sustained heightened subcortical metabolic activity, a premorbid vulnerability marker that, together with a developmental reward-control imbalance, could contribute to adolescent risk for early substance use initiation before behavioral symptoms emerge. Characterization of such developmental patterns is critical for informing the search for biomarkers of risk for hazardous behaviors during adolescence, including initiation of alcohol and substance use and mood and anxiety symptoms that increasingly emerge during the second decade of life.

Keywords: Adolescence, Cerebral Blood Flow, Arterial Spin Labeling, Substance Use Initiation, Reward Circuitry

Disclosure: Nothing to disclose.

P4. SEX HORMONE-RELATED HIPPOCAMPAL DEVELOPMENT DURING PUBERTY: LONGITUDINAL EVIDENCE FROM

REPEATED CROSS-SECTIONAL AND CUMULATIVE ESTRADIOL AND TESTOSTERONE MEASURES

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Background: Puberty is a critical developmental window marked by significant changes in behavior, emotion, and cognition, as well as the emergence of well-documented sex differences in neuropsychiatric disorders. Preclinical and human evidence has shown that the hippocampus undergoes significant structural and functional remodeling during this period, and that pubertal gonadal hormone secretion directly influences hippocampal morphology. In humans, associations between sex hormones and hippocampal structure have been reported, as have been the emergence of sex differences, but findings are often confounded by factors such as age, menstrual cycle status, reliance on cross-sectional designs, or infrequent longitudinal sampling during this phase of rapid neurodevelopment. Here, using data from the ongoing "NIMH Intramural Longitudinal Study of the Endocrine and Neurobiological Events Accompanying Puberty," we characterized developmental trajectories of hippocampal volume alongside plasma estradiol and testosterone levels in healthy boys and girls from prepuberty through age 18 to investigate possible hormone-related patterns of change.

Methods: Healthy children ($N = 135$; 56 girls) between the ages of 8 and 18 years were assessed every 9 months with fasting morning blood draws and 3T structural MRI (GE MR750, 1 mm isotropic voxels; $TE = 1.8$ ms; $TR = 10.5$ ms), totaling 566 visits. Prepubertal status at age 8 and pubertal stage at every 9-month visit thereafter were ascertained by clinicians. To control for menstrual cycle-related hormonal effects, structural scans in post-menarcheal girls were collected in the follicular phase (menstrual cycle days 4–11) and confirmed by menses dates and plasma progesterone < 2 ng/ml. Hippocampal volumes were derived using FreeSurfer. Analyses included repeated-measures correlations (R : r_{mcorr}) and longitudinal mixed-effects spline models (R : gam4) correlating hippocampal volume to repeated cross-sectional versus cumulative hormone measures. Cross-sectional hormone levels were defined as plasma estradiol and testosterone levels measured at each visit, whereas cumulative exposure was quantified as the area under the longitudinal hormone secretion curve, reflecting both absolute levels and duration of exposure.

Results: Plasma estradiol and testosterone levels were positively correlated with bilateral hippocampal volumes in both sexes irrespective of age (all p 's < 0.0006 , r 's > 0.22). Longitudinal spline models of cross-sectional estradiol levels revealed significant age-by-estradiol interactions in the bilateral hippocampus in girls such that those with higher estradiol levels exhibited a steeper age-related change in hippocampal volume than girls with lower estradiol levels ($p < 0.001$). In contrast, cross-sectional testosterone levels in boys were not significantly associated with age-related hippocampal changes. However, cumulative testosterone exposure across puberty significantly predicted hippocampal volume such that boys with lower cumulative testosterone levels showed a steeper age-related change in hippocampal volume than boys with higher cumulative testosterone levels ($p = 0.002$), consistent with some (but not all) animal and human studies.

Conclusions: Our longitudinal findings empirically demonstrate estradiol- and testosterone-dependent hippocampal structural changes across puberty. Hormone-associated variations in hippocampal development during this sensitive period may contribute to the emergence of sex differences and increased vulnerability to neuropsychiatric disorders that are well documented during this

period. Importantly, our findings highlight the need to consider cumulative hormone exposure, rather than relying solely on cross-sectional measures, to more accurately capture the developmental impact of pubertal hormones on the brain. Future work will additionally evaluate the effects of cumulative estradiol exposure, as well as of bone age, a proxy for cumulative estradiol exposure, in boys and girls, on hippocampal development.

Keywords: Puberty, 17- β -estradiol, testosterone, Brain development, Children and Adolescents

Disclosure: Nothing to disclose.

P5. MAPPING INTRA AND INTER-TASK RELATIONSHIPS: VALENCE FLEXIBILITY AND AMYGDALA ACTIVATION IN HEALTHY ADOLESCENTS

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Background: Adolescence is marked by profound cognitive and emotional change. While cognitive flexibility has been studied in development, our understanding of flexibility in the emotional domain remains limited. The ability to efficiently shift between negative and positive emotional states, i.e., valence flexibility, may play a critical role in normative development; whereas, difficulties in flexibly switching between emotional valences may contribute to the onset of psychopathology. Investigating valence flexibility across various levels of emotional processing (i.e., emotion recognition, evocation, and appraisal) could offer valuable clinical insights.

Methods: In a 3T MRI scanner, a healthy sample of youth ($n = 48$, 9–17 years old, 13.75 ± 2.66 years) completed multiple valence flexibility tasks in the scanner in randomized order. In the emotion recognition task, individuals viewed two oppositely-valenced facial expressions (e.g., happy, angry) simultaneously and located the face that matched a central word cue (e.g., angry). Individuals viewed happy-angry and happy-sad pairs. The position of the negative face was counterbalanced across trials. In the emotion evocation task, participants viewed emotional images from the International Affective Picture System and rated whether each image evoked positive or negative feelings. In the emotion appraisal task, individuals indicated whether statements were true or false about themselves (i.e., self-appraisal) or a same-sex peer (i.e., other appraisal). In one condition, the statements were mixed (i.e., mixed appraisal). In all tasks, the valence on consecutive trials switched (e.g., negative-positive) or repeated (e.g., negative-negative). Reaction times (RT) were recorded via button-press.

Switch cost for each task was calculated as the difference between switch and repeat trials, with greater switch costs reflecting inflexibility. Behaviorally, RT differences were calculated after removing inaccurate trials and outliers. For neuroimaging data, individual models were created for each task separately. Percent signal change was extracted from the amygdala using an anatomical mask. Amygdala activation in response to switch cost was calculated. Correlations within and between tasks were conducted using the switch cost contrast (i.e., switch vs. repeat). Statistical significance was determined using an $\alpha = 0.05$ threshold.

Results: For both happy-angry and happy sad blocks of the emotion recognition task, RT for switch was slower than repeat trials ($t(46) > 2.4$, both $p < 0.021$), but RT for switch trials were faster than repeat trials in the mixed block of the emotion appraisal task ($t(47) > -2.3$, $p < 0.024$). Right amygdala activation in response to switch vs. repeat was observed only during emotion recognition of happy-angry ($t(31)$, $t(34) = 2.3$, $p < 0.028$). No other effects were detected (all $p > 0.14$).

We examined the relationships between different conditions within the emotion recognition (i.e., happy-angry, happy-sad) and the emotion appraisal tasks (i.e., self, other, and mixed). These analyses did not apply to emotion evocation. No relationships were detected in switch cost based on RT (all $p > 0.3$). The switch cost in left amygdala activation to happy-angry blocks and happy-sad emotion recognition blocks were positively correlated ($r(32) = 0.415$, $p < 0.018$), but not the right amygdala (all $p > 0.2$). The switch cost in right amygdala activation in the self was negatively correlated with the other appraisal block ($r(37) = -0.573$, $p = 0.001$). No other effects were noted (all $p > 0.12$).

Finally, we examined brain relationships across recognition, evocation and appraisal tasks. In terms of RT differences, the switch cost between emotion evocation and emotion appraisal of self was positively related ($r(47) = 0.40$, $p < 0.006$). In left amygdala, switch cost in activation between emotion evocation was negatively associated when appraising others ($r(37) = -0.337$, $p < 0.041$). In the right amygdala, switch cost in activation between emotion evocation was positively associated with appraising others ($r(37) = 0.432$, $p < 0.008$) and in mixed blocks ($r(37) = 0.334$, $p < 0.043$).

Of note, Screen for Child Anxiety-related Disorders (SCARED, Mean = 17.6, SD = 11.6) was not associated with switch cost based on RT or amygdala activation in any task, all $p > 0.08$.

Conclusions: Several findings should be noted. In the emotion recognition task, both RT and right amygdala activation was significantly greater in switch than repeated trials. In addition, left amygdala activation was positively correlated between negative conditions of the emotion recognition task, suggesting emotion recognition may be more stable and less susceptible to change. Interestingly, in terms of right amygdala activation, greater inflexibility (i.e., switch > repeat) to self was associated with more amygdala flexibility (i.e., repeat > switch) to others. This inverse relationship may suggest flexibility may be applied more easily to others than oneself. Finally, switch cost to emotion evocation was related to switch cost in other levels of emotional processing (e.g., self-appraisal), but the direction varied depending on the appraisal type and hemisphere. These findings underscore the complexity of valence flexibility across emotional domains and suggests that individual differences in behavioral and neural flexibility may have implications for the development of socio-emotional functioning and mental health.

Funding: R21MH112928 (JCB)

Keywords: Facial emotion processing, emotional processing, Amygdala, Anxiety, Adolescent

Disclosure: Nothing to disclose.

P6. WHITE MATTER, FAMILY ENVIRONMENT, IMPULSIVITY, AND SEX AS PREDICTORS OF NON-SUICIDAL SELF-INJURY AND CONCURRENT SUICIDAL THOUGHTS AND BEHAVIORS IN THE ADOLESCENT BRAIN COGNITIVE DEVELOPMENT STUDY

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Background: Non-suicidal self-injury (NSSI) often begins in adolescence and is linked to increased suicide risk. Diffusion weighted imaging (DWI) studies suggest altered white matter in youth with NSSI in the cingulum and uncinate fasciculus (UF). Other associated factors include family dynamics, impulsivity, and sex. Existing research is limited by small samples, few male

participants, cross-sectional designs, and lack of clinically matched controls. We used data from the Adolescent Brain Cognitive Development (ABCD) study to address these gaps. We examined whether baseline white matter microstructure, family conflict/support, impulsivity, and sex predicted NSSI onset two years later (Y2).

Methods: Participants were 418 youth (ages 8–11 years, 66% female); 209 who endorsed first onset NSSI at Y2, and 209 non-NSSI clinically matched controls. Baseline predictor variables of interest included generalized fractional anisotropy (GFA) from the bilateral UF and six segments of the cingulum, Family Conflict from the Family Environment Scale (FES), and Parental Acceptance from the Child's Report of Parent Behavior Inventory (CRPBI). We also used the Positive and Negative Urgency, Premeditation, Perseverance, and Sensation Seeking impulsivity subscales of the UPPS-P. NSSI and STB were measured at baseline and at Y2 using the KSADS. Within the NSSI group, 108 endorsed Y2 STB. For each analysis (1. NSSI and 2. STB), we first completed univariate analyses for each predictor. Predictors with $p < 0.25$ in univariate analyses were included in the final logistic regressions along with sex and interaction with sex. Intracranial volume and MRI manufacturer were included as covariates.

Results: For NSSI, there were main effects in which lower GFA of the left cingulum (parolfactory region; OR = 0.46, $p < 0.01$) and higher Negative Urgency (OR = 1.16, $p = 0.04$) at baseline were associated with higher odds of Y2 NSSI. There were significant interactions with sex as females showed opposing patterns (OR = 2.45 and 0.83 for GFA and Negative Urgency respectively). Further, higher Sensation Seeking in females was associated with higher odds of Y2 NSSI (OR = 1.20, $p = 0.04$). For STBs within the NSSI group, females had significantly higher odds of concurrent Y2 STB (OR > 5, $p = 0.04$).

Conclusions: White matter GFA and impulsivity at baseline predict NSSI onset two years later, with males showing a particularly strong relation between Y2 NSSI and baseline GFA and tendency to act rashly in the context of negative emotion. Among youth with NSSI, females were significantly more likely to endorse concurrent STB. The sex differences highlighted by this study emphasizes the importance of tailoring intervention and prevention approaches accordingly. Additional analyses examining males and females separately are warranted given the lack of research in this area, particularly for males.

Keywords: Adolescence, Non-Suicidal Self-injury (NSSI), suicide, diffusion MRI, ABCD

Disclosure: Nothing to disclose.

P7. A RISK CALCULATOR FOR PREDICTING SUBSEQUENT-YEAR PSYCHOTIC EXPERIENCES IN EARLY ADOLESCENCE

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Background: Persistent psychotic experiences during adolescence have been associated with an increased likelihood of developing a psychiatric disorder in adulthood, including but not limited to psychotic disorders. Psychotic experiences encompass unusual thoughts or perceptions, reality confusions, disorganized speech, and delusions of persecution or unusual abilities. Transient experiences are relatively common in late childhood and adolescence and often benign, but risk factors for persistence have been identified. Building upon existing risk calculators that focus on older, help-seeking populations with more advanced illness, we aimed to estimate the likelihood of subsequent-year psychotic symptoms in a community sample of young adolescents.

Methods: Our prediction model uses five years of data from the longitudinal Adolescent Brain Cognitive Development Study, which enrolled over 11,000 nine- and ten-year-olds across 21 sites in the United States. Yearly self-reports of psychotic experiences were collected using the Prodromal Questionnaire-Brief Child Version (PQ-BC) and severity scores were divided into three symptom states (low, medium, high) based on prior literature. Features used to predict the current symptom state included prior-year symptom state, sex, static risk factors (adverse childhood experiences (ACEs), perinatal adversity, psychiatric family history, developmental delay), dynamic risk factors (bullying, stressful life experiences, alcohol experimentation), and psychiatric/behavioral indicators for concern (internalizing, externalizing and attention problems, self-harm, school problems, mental healthcare, social isolation). Only participants who completed the PQ-BC were included at a given timepoint (n participants = 9785, n observations = 30,027) and the cohort was split into sociodemographically-matched discovery and replication sets. All other features were imputed when missing, and numeric variables were scaled. Model selection across regularized logistic regression, tree-based models, and support vector machines was conducted in the discovery cohort using 5-fold cross-validation with hyperparameter tuning. The model with the highest average balanced accuracy across folds was selected. To assess generalizability, performance on the replication cohort was evaluated with balanced accuracy, per-class recall, and both a per-class and a multiclass metric of the area under the receiver operating characteristic curve (ROC-AUC). To interpret model behavior, explainability methods were performed.

Results: Regularized logistic regression performed best across discovery cohort cross-validation (balanced accuracy = 0.538 ± 0.013). Comparable performance was achieved with a random forest classifier (balanced accuracy = 0.524 ± 0.010) and non-linear kernel support vector machines (balanced accuracy = 0.519 ± 0.016). The best model generalized well to the replication cohort (balanced accuracy = 0.521; per-class recall = 0.783 (low), 0.259 (medium), 0.522 (high); multiclass ROC-AUC = 0.76; per-class ROC-AUC = 0.79 (low), 0.67 (medium), 0.82 (high). Permutation- and SHAP (SHapley Additive exPlanations)-based feature importance analyses revealed previous symptom state, internalizing problems, and attention problems as the most important predictors. Across all models, ACEs and stressful life events were also consistently ranked as important.

Conclusions: Performance and generalizability in the replication cohort suggest that this risk calculator could be a useful clinical tool for identifying adolescents at risk for later psychiatric disorders based on their likelihood of having persistent psychotic experiences. We plan to extend this calculator across future years of the ABCD Study to examine how model performance and key predictors evolve throughout adolescence, and to assess whether individuals predicted to be at risk ultimately develop psychiatric disorders.

Keywords: adolescent development, machine learning, psychotic-like experiences, Risk Calculator

Disclosure: Nothing to disclose.

P8. BASAL GANGLIA IRON CONTENT AND MYELINATION IN ADOLESCENTS

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Background: Brain iron is essential for many functions, including myelin production and maintenance. Yet, iron deficiency (ID) remains the most common micronutrient deficiency, affecting 38.6% of female adolescents in the US. We have recently shown

that ID without anemia was associated with reduced basal ganglia (BG) iron content in female adolescents and poorer psychiatric and cognitive functioning. Here, we examined the association between BG iron content and neuroimaging-based microstructural properties in adolescents.

Methods: Unmedicated 10- to 17-year-old participants, with and without internalizing disorders, were enrolled in a cross-sectional magnetic resonance imaging (MRI) study, including structural (T1w and T2w) MRI, diffusion tensor imaging (DTI), and quantitative susceptibility mapping (QSM). Major white matter fiber maturation was indexed by fractional anisotropy (FA) calculated from DTI. T1w/T2w ratio reflects relative myelin content in white matter and a combined contrast of iron and myelin in gray matter. The association of BG susceptibility, quantified from QSM, with FA and T1w/T2w ratio was examined using tract-based spatial statistics (TBSS) and voxel-based statistics analyses, respectively, controlling for age.

Results: Two hundred and six adolescents (58% female, age: 13.6 ± 2.2 years) provided usable data. After adjusting for age, putamen susceptibility was significantly ($p < 0.05$) positively associated with FA in the superior longitudinal fasciculus and external capsule, which was corroborated by the T1w/T2w ratio with additional associations across temporal and cerebellar cortices as well as the striatum. None of these results was significant ($p < 0.05$) in male adolescents.

Conclusions: Cortical and white matter maturation in female adolescents is associated with basal ganglia iron content, which itself is reduced in ID, even in the absence of anemia. The lack of such association in male adolescents parallels a similar lack of reduced BG iron content in male adolescents with ID in our recent publication, which further highlights the priority of ID research in females. Our findings provide new insights into the microstructural underpinnings whereby ID may disrupt brain development and contribute to psychopathology, requiring urgent preventive interventions.

Keywords: Adolescence- Critical Period, Adolescent Depression, Adolescent Anxiety, Brain development

Disclosure: Nothing to disclose.

P9. TIMECOURSE OF DIGITAL EMOTION ASSESSMENT VARIES IN RELATIONSHIP TO ADOLESCENT PSYCHOPATHOLOGY

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Background: Standard clinical measures of affect provide only a static view of the emotional experiences of youth with psychopathology when affect is dynamic, changing momentarily, day-to-day, and even week-to-week. While prior research has shown that overall higher negative affect and lower positive affect characterizes multiple psychiatric disorders with developmental origins, including major depressive disorder (MDD) and borderline personality disorder (BPD), less is known about whether the variability in these emotional experiences is similarly related MDD and BPD.

Methods: Here we use ecological momentary assessment to the relationship between both overall affect and variability in affect with MDD and BPD symptoms in 57 adolescents (ages 10–15 years, mean 12.9) enrolled in a longitudinal study of early onset depression who reported on negative and positive affect four times per day for a week, daily for 30 days, and weekly for one year. We examined associations between (a) mean levels of and (b) variability of positive and negative affect and symptoms and

diagnoses of MDD and BPD and whether these associations differed based on timescale.

Results: Higher negative affect and lower positive affect was correlated with depressive symptoms and MDD diagnosis across all timescales (momentary, daily, weekly). Greater momentary negative affect and its variability was also correlated with depressive symptoms, though not MDD diagnosis. Higher negative affect and lower positive affect was also correlated with BPD symptoms across all timescales, and both momentary and day-to-day variability in positive and negative affect was also correlated with BPD diagnosis. However, variability in positive affect was uniquely associated with BPD.

Conclusions: Using digital EMA, there are similar patterns between ratings of overall positive and negative affect with MDD and BPD symptoms, but variability in positive affect shows a unique association with BPD. Thus, frequent digital EMA sampling may be important in accurately studying BPD and provide an opportunity to differentiate between clinical disorders with overlapping symptoms.

Keywords: Borderline Personality, Adolescent Depression, ecological momentary assessment

Disclosure: Nothing to disclose.

P10. LONGITUDINAL [18F] SYNVEST-1 PET IMAGING REVEALS REGION-SPECIFIC SYNAPTIC REDUCTIONS IN MENTAL HEALTH SEEKING YOUTH: PRELIMINARY EVIDENCE FROM THE TAY COHORT STUDY

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Background: Synaptic pruning during adolescence is essential for healthy brain development. Disruptions in this process are hypothesized to underlie vulnerability to psychiatric disorders. PET imaging of synaptic vesicle glycoprotein 2A (SV2A) provides an in vivo measure of synaptic density, offering insights into neurodevelopmental trajectories in mental illness. The aim of this study was to longitudinally characterize synaptic density in mental health treatment-seeking youth using [18F]SynVesT-1 PET imaging. We hypothesized that synaptic density would decline over time reflecting synaptic pruning.

Methods: Participants aged 16 and older from the Toronto Adolescent and Youth (TAY) Cohort Study - an ongoing longitudinal cohort study aiming to collect multimodal data in 1500 11–24 year-olds - underwent two [18F]SynVesT-1 PET scans approximately one year apart, along with structural MRI for region-of-interest definition. Twenty-four treatment-seeking youth (13 male, 11 female; mean age 20) were included. Non-displaceable binding potential (BPnd) was estimated using a simplified reference tissue model. Longitudinal changes were assessed using a repeated measures ANOVA.

Results: [18F]SynVesT-1 BPnd significantly declined across the brain ($F(1, 23)8.876$; $p = 0.007$), with region-specific differences ($F(5,115)12.37$; $p < 0.001$): hippocampus (-13.4% ; $p = p < 0.001$), amygdala (-10.7% ; $p = p < 0.001$), striatum (-8.7% ; $p = p < 0.007$) and anterior cingulate cortex (-11.7% ; $p = p < 0.003$) but not prefrontal and temporal cortices (-3% ; ns). Youth with psychosis spectrum symptoms (based on PRIME Screen-Revised (PRIME)) showed greater hippocampal (-17% vs. -6%) and amygdala (-15% vs. -1%) reductions compared to those without PSS.

Participants with PTSD also showed greater hippocampal decline (-18% vs. -7%).

Conclusions: Preliminary longitudinal data suggest that synaptic pruning, as measured by SV2A PET, continues into late adolescence and early adulthood, with greater reductions observed in limbic compared to cortical regions. Exploratory analyses indicate that synaptic density loss may be more pronounced in treatment-seeking youth with psychosis-spectrum symptoms and posttraumatic stress, highlighting regionally selective effects that may underlie stress-related psychopathology during this critical developmental period.

Keywords: Synaptic Function, PET, Adolescence, Adolescent PTSD, Psychosis spectrum symptoms

Disclosure: Nothing to disclose.

P11. NEURAL CORRELATES OF REWARD DYSFUNCTION IN ADOLESCENT CANNABIS USE AND DEPRESSION

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Background: Cannabis has surpassed alcohol as the most used illicit substance among U.S. adolescents since the early 2010s, and its use is more than twice as prevalent among those with depression. Preclinical studies suggest that juvenile exposure to $\Delta 9$ -tetrahydrocannabinol (THC), the psychoactive agent in cannabis, could irreversibly perturb reward regions, including the striatum and prefrontal cortex. Echoing these findings, the few existing neuroimaging studies on adolescent cannabis use have documented structural and functional alterations within the reward circuitry, a system also central to the neurobiology of depression. However, research directly examining cannabis use and depression together in adolescence remains limited. Addressing this gap, we sought to investigate reward function in a sample of adolescents presenting across a range of cannabis use frequency and depression severity using the fMRI Reward Flanker Task (RFT).

Methods: Adolescents from the New York Metro area had semi-structured diagnostic interviews with trained clinicians. Depression severity was measured using the clinician-rated Children's Depression Rating Scale-Revised. Cannabis use patterns were assessed via clinician interviews, self-reports, and urine toxicology. Use frequency was defined by clinician consensus as none, tried once, low, moderate, or heavy. Structural MRI (T1w MPRAGE and T2w SPACE; 0.9 mm isotropic) and RFT fMRI (2.3 mm isotropic; TR = 1s) were acquired and processed using Human Connectome Project-style methods, including ICA-FIX denoising and MSMALL alignment. RFT trials presented a monetary cue (0, 10, 50, or ? cents), a brief flanker stimulus (unique letter among 5), a window to identify the unique letter, and feedback on accuracy and reward. The RFT included 120 trials over 4 runs. Neural activation during reward expectancy (reward vs. non-reward cues) and attainment (reward vs. non-reward feedback) were modeled in SPM. Whole-brain data were parcellated using the Cole-Anticevic Brain-wide Network Partition. Group-level non-parametric analyses were conducted in FSL PALM (10,000 permutations), controlling for familywise error (FWE), age, and sex. Reward network masks, comprising the top 10% of nodes activated by reward expectancy and attainment contrasts identified in a prior study, were applied. Cannabis use and depression severity were modeled together, such that each effect reflected associations independent of the other. If there was a significant main effect, interaction effects were subsequently examined. Secondary analyses explored sex differences. This parcellated, network-focused approach balanced neurobiological detail with statistical

power. Effect sizes (ES) were computed from t-values and degrees of freedom for all analyses, except the sex-stratified analyses due to their exploratory nature. All results reported were significant at $p_{FWE} < 0.05$.

Results: The sample included 117 adolescents (15.5 ± 2.3 years, 63.3% female) with usable fMRI data. Across all participants, greater depression severity was associated with blunted left caudate activation during reward expectancy ($ES = -0.340$) and heightened left posterior cingulate cortex (PCC; $ES = 0.363$) and entorhinal cortex (EC; $ES = 0.323$) activation during reward attainment. In those with cannabis use ($n = 32$), heavier use was linked to greater activation in the parafascicular thalamic nuclei, nucleus limitans, and habenula during reward expectancy ($ES = 0.660$). In this group, depression severity also related to widespread hyperactivation during reward attainment in the left nucleus accumbens, PCC, EC, dorsolateral prefrontal cortex, inferior temporal gyrus (ITG), anterior cingulate cortex, visual cortex, right rostral prefrontal cortex, and bilateral precuneus, with ES ranging from 0.612 to 0.746. A significant Cannabis Use $\times$ Depression interaction emerged during reward attainment; in the full sample, the interaction was associated with hyperactivation in the left ITG ($ES = 0.335$) and frontal pole ($ES = 0.353$), while in those who used cannabis, the interaction was positive in the left EC ($ES = 0.621$). Notable sex-specific effects were: (a) in males across the sample ($n = 44$), depression severity correlated negatively with left caudate activation during reward expectancy, and (b) in females who used cannabis ($n = 23$), more frequent use was linked to greater left mediodorsal thalamus and habenula activation during reward expectancy.

Conclusions: In the context of comorbidity, heavier cannabis use may be linked to altered neural function during reward expectancy, engaging areas important to both reward and anti-reward processing. In contrast, more severe depression may involve heightened and distributed neural engagement during reward receipt, recruiting self-referential, executive control, memory, and visual processes. Future studies with larger cohorts and longitudinal designs are needed to further clarify the trajectory of reward dysfunction in co-occurring cannabis use and depression among adolescents.

Keywords: cannabis use, depression, task fMRI, reward, Adolescence

Disclosure: Nothing to disclose.

P12. TOWARD PRECISION PREDICTION OF ADOLESCENT DEPRESSION USING MULTIMODAL NEUROIMAGING AND ELASTIC NET REGRESSION

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Background: Adolescent depression is a prevalent and impairing psychiatric condition, yet predicting depressive symptom severity at the individual level remains a major challenge. The multifactorial nature of depression risk, along with the distributed organization of brain function, contributes to this difficulty. Recent advances in neuroimaging have enabled the extraction of hundreds of features across structural, functional, and diffusion modalities. However, integrating such high-dimensional data to predict individual symptom severity in large samples remains underdeveloped. Multivariate machine learning approaches such as elastic net regression, paired with dimensionality reduction techniques like principal component analysis (PCA), provide a promising avenue to improve individualized prediction. In this

study, we sought to determine whether multimodal neuroimaging data could be used to predict depressive symptoms in adolescents, while accounting for site and demographic variation. We hypothesized that distributed features related to emotion regulation, salience, and cognitive control would be among the most predictive of depressive symptom severity.

Methods: We analyzed data from 5769 participants in the Adolescent Brain Cognitive Development (ABCD) study. A total of 560 neuroimaging features spanning cortical thickness, surface area, subcortical volume, resting-state connectivity, task activation (Monetary Incentive Delay and Stop Signal Tasks), and diffusion metrics were reduced via PCA to 50 orthogonal components. These components were then used in an elastic net regression model with 5-fold cross-validation to predict CBCL DSM-5 Depression T-scores. Covariates included age, sex, race/ethnicity, household income, intracranial volume, and motion parameters. Scanner site effects were adjusted using linear mixed-effects modeling. Model performance was evaluated using root mean squared error, mean absolute error, mean squared error, and R-squared.

Results: Our final model demonstrated modest but reliable prediction of depressive symptom severity using neuroimaging-derived components and covariates. The most informative predictors were drawn from brain activation during the Monetary Incentive Delay (reward anticipation) and Stop Signal (inhibitory control) tasks. Brain regions contributing most strongly included the insula, inferior frontal gyrus, precentral gyrus, thalamus, and brainstem—regions involved in interoception, motor control, and emotional regulation. Notably, measures of signal stability (e.g., standard errors of activation estimates) contributed significantly, underscoring the importance of both the strength and consistency of neural responses. These findings suggest that predictive value arises not only from regional activation but also from the reliability of that activity over time.

Conclusions: This study highlights the potential of multimodal neuroimaging data, when distilled into orthogonal components and modeled using elastic net regression, to yield modest yet interpretable predictions of adolescent depressive symptoms. The implicated networks—interoceptive-affective, salience, and motor control—align with known neurobiological models of depression, emphasizing dysregulation across cognitive and emotional systems. Although model performance remains limited, these results support the utility of machine learning pipelines incorporating dimensionality reduction and regularization to explore precision psychiatry in youth. Future efforts integrating longitudinal imaging, psychosocial risk factors, and ecological assessments may enhance predictive accuracy and clinical relevance.

Keywords: Adolescent Depression, ABCD study, machine learning, Multimodal Neuroimaging, Brain-Based Predictive Modeling

Disclosure: Nothing to disclose.

P13. DEVELOPMENTAL ASPECTS OF THE PROSPECT THEORY - WHAT WE CAN LEARN FROM THE ABCD STUDY

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Background: Prospect Theory, developed by Kahneman and Tversky, transformed economic and psychological models of decision-making by showing that humans systematically deviate from rational, utility-based frameworks. A core principle is loss aversion: adults consistently weigh losses more heavily than

equivalent gains, becoming risk-averse in gain contexts but more risk-seeking under potential loss. This asymmetry has been robustly demonstrated across monetary and non-monetary domains, underscoring its pervasive role in shaping real-world behavior.

Despite this strong evidence in adults, far less is known about whether Prospect Theory adequately describes how young people approach risky prospects. Late childhood and early adolescence represent critical neurodevelopmental stages marked by changes in cognitive control, affect regulation, and abstract reasoning, all of which may influence the emergence of decision-making heuristics. Understanding whether risk-taking patterns in gain versus loss contexts reflect constitutional traits or instead emerge through socialization and developmental experience represents a novel extension of Prospect Theory. Developmental investigation of Prospect Theory may identify sensitive periods when valuation biases consolidate, informing education, financial literacy, prevention of maladaptive risk behaviors, as well as characterizing the onset of conditions where reward sensitivity and risk processing are often disrupted (e.g., ADHD, depression, anxiety, and substance use). Correlating Prospect Theory parameters with impulsivity and behavioral outcomes, as measured by the UPPS and CBCL, respectively, provides a bridge to clinically relevant phenotypes. Furthermore, linking these behavioral indices to neural development—particularly neurite density—offers mechanistic insight into how cognitive biases interface with brain maturation.

In this study, we leveraged the nationally representative ABCD Study to examine developmental patterns of gain–loss decision-making, their behavioral correlates, and associated neural architecture.

Methods: From the ABCD Study, 8,666 ($M = 52\%$) participants from baseline (ages 9–10 years) who successfully performed the Monetary Incentive Delay (MID) task were included. The MID task is designed to assess anticipation and outcome of gain or loss during five possible trial types [Small Win \$.20, Large Win (\$5), Small Lose (\$.20), Large Lose (\$5), and neutral (\$0)]. All participants underwent extensive baseline assessments, including the CBCL and UPPS questionnaires. Using the reinforcement learning framework, we identified probability of gain (PoG) as the behavioral index of rewarded decision making. To investigate the principle is loss aversion, compared PoG between task loss and reward task conditions. We further stratified the groups into quartiles based on their PoG for large losses and separately for large rewards and examined five subgroups: (1) Large-Loss/Small-Reward, (2) Small-Loss/Large-Reward, (3) Large-Loss/Small-Reward, (4) Large-Loss/Large-Reward, and (5) Controls (mid 50% on loss and reward). Linear mixed-effects models were used to examine between-group differences in UPPS subscores, CBCL scores, and neurite density of regions implicated in reward processing.

Results: Across all participants, PoG was the lowest for Large-Reward and the highest for Large-Loss ($t = -19.39$, $p < 0.001$) with PoG for Small-Reward and Small-Loss being in-between. Contrasts between each pair of variables were highly significant ($p < 0.001$). Between group comparisons showed that compared to Controls, the Large-Loss/Small-Reward group showed significantly higher impulsivity (negative urgency and sensation seeking; $p\text{-FDR} < 0.01$), as well as aggression ($p\text{-FDR} = 0.02$) and externalizing behaviors ($p\text{-FDR} = 0.016$). For neurite density, compared to Controls, the Large-Loss/Small-Reward group showed higher neurite density in the bilateral caudate and putamen ($p\text{-FDR} < 0.009$), accumbens ($p\text{-FDR} < 0.02$), hippocampus ($p\text{-FDR} < 0.001$), as well as lateral ($p\text{-FDR} < 0.05$) and medial ($p\text{-FDR} < 0.04$) orbitofrontal cortices. Mediation and moderation analyses are currently underway.

Conclusions: These findings demonstrate that adolescents showing heightened loss-related decision-making also exhibit

greater impulsivity, aggression, and externalizing behaviors, paralleling alterations in striato-cortical neurite density. Elevated neurite density in these regions is consistent with prior evidence suggesting delayed synaptic pruning in adolescence, reflecting less mature reward circuitry. Together, these results suggest that developmental variations in Prospect Theory parameters may capture vulnerability to externalizing problems through both behavioral and neural pathways.

Keywords: Prospect theory, Reward sensitivity, Impulsivity, adolescence, ABCD study

Disclosure: Nothing to disclose.

P14. ADOLESCENT CHRONIC STRESS ALTERS SYNAPTIC TRANSMISSION AND NEURONAL ACTIVATION IN LAYER 2/3 OF THE PRELIMBIC CORTEX

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Background: Early Life Stress (ELS), such as childhood neglect and trauma, is a key risk factor for psychiatric disorders like PTSD and major depression, often leading to deficits in social processing. Two-thirds of U.S. youth experience trauma by adolescence, a developmental stage when neural, biological, and psychosocial systems are maturing and highly sensitive to external influences like social interactions. When these interactions are traumatic, they can alter the trajectory of the development of neurocognitive processes crucial for decision-making and other executive functions that lead to maladaptive behaviors that persist to adulthood. The prefrontal cortex (PFC) undergoes prolonged maturation, which is critical for its role in top-down regulation and decision-making. This extended development makes the PFC vulnerable to adolescent chronic stress. The mechanistic links between stress during adolescence and long-term social dysfunction remains unknown, making informed early intervention efforts limited. Here, we use a rodent model to explore immediate and long-term changes in neural activation and synaptic transmission in the prefrontal cortex after chronic adolescent stress (chronic social defeat stress, CSDS).

Methods: FosTRAP2 (FosCreER; Ai14) male mice, which allow for permanent labeling of neurons active during an event such as stress or social behavior, were used for adolescence (postnatal day (PND) 28–37) CSDS (aCSDS) experiments. Briefly, the FosTRAP2-aCSDS group mice were exposed to a CFW aggressor for 30 bites or 5 min, then co-housed and separated by a perforated partition. This was repeated daily for 10 days with different aggressors. FosTRAP2-controls were pair-housed with a perforated partition. Cells activated during aCSDS were tagged by injecting 4-hydroxytamoxifen (4-OHT, 50 mg/kg) via intraperitoneal injection, 60 minutes after defeat on multiple days. Open field social interaction (OFSI) with a non-aggressive CFW was done 24 h after CSDS. Mice were transcardially perfused 2 hours after OFSI, and brains were sectioned at 20 μm for immunofluorescent labeling of Fos induced by the OFSI. QuPath software was used to count the number of FosTRAP and Fos-positive cells per area. For electrophysiological experiments, whole-cell patch-clamp recordings were performed in slices from adult mice (>PND70). Spontaneous inhibitory postsynaptic currents (IPSCs) and excitatory postsynaptic currents (eEPSCs) were recorded from deep layer 2/3 prefrontal cortex (PL) neurons in the same neurons at holding potentials of 0 mV or -70 mV, respectively.

Results: FosTRAP-aCSDS mice display a social avoidance phenotype and reduction in body weight gain. Adult FosTRAP-aCSDS show

increase in the amplitude of sIPSCs in prelimbic layer 2/3 neurons activated during aCSDS compared to controls ($N = 10\text{--}12$ neurons from 4 to 5 mice, $p = 0.05$; student t -test). We did not observe this phenotype in the Fos negative (not activated by aCSDS) cells. Preliminary data show that adolescent FosTRAP-aCSDS mice have a population of cells activated during defeat in Layer 2/3 of the PL and a subpopulation of these cells reactivated during subsequent social interaction ($N = 6$ sections per animal, 2–5 per group).

Conclusions: Our findings suggest that aCSDS may lead to the imbalance in excitation/inhibition ratio in a subpopulation of layer 2/3 neurons in the PL which may contribute to social deficits. The identification of cells reactivated during OFSI after aCSDS could provide insight into the regulation of social behaviors. Future studies will target these cells to mitigate or prevent the long-lasting effects of observed dysfunctions on social behaviors.

Keywords: Chronic adolescence stress, Medial Prefrontal Cortex (mPFC), Postsynaptic transmission, Social interaction, Neural activation

Disclosure: Nothing to disclose.

P15. NEUROCOGNITIVE, GENETIC AND SOCIOENVIRONMENTAL INFLUENCES ON DEVELOPMENTAL PRECURSORS TO SUBSTANCE USE: A CROSS-SPECIES STUDY

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Background: Early environmental and genetic influences exert significant effects on risk for Substance Use Disorders (SUDs). This abstract describes progress from a bidirectional translational team performing a NIDA-funded R34 project aimed at identifying both behavioral and genetic risk factors for neurocognitive phenotypes that precede SUDs. We are employing a cross-species study that includes: (1) determining the extent to which early life environment influences neurocognitive development via adult delay-discounting behavior in laboratory rats; (2) identifying transcriptional network signatures of early life adversity (ELA) from publicly available datasets of rats and mice; and (3) generating polygenic risk scores (PRS) for externalizing-related phenotypes from human GWAS datasets to assess the extent to which these PRS influence key behavioral outcomes (e.g., impulsivity) thought to be precursors to addiction in The Adolescent Brain and Cognitive Development (ABCD) Study.

Methods: For (1), male and female Long Evans rats on postnatal days (PND) 2–21 were subjected to low, standard, or high resource housing conditions, which ranged in bedding volume and material, cage size, and environmental enrichment. We are generating final groups of 4/sex/litter with a total of 10 litters/housing condition; but are currently powered at 6–10 rats/sex/condition. On PND 21–75, we collected developmentally relevant behavior. On PND 75, the rats began operant training for the delay discounting procedure. Training progressed through successive steps of responding on one lever for one 32% liquid food delivery and responding on the other lever for four 32% liquid food deliveries. Following acquisition, delay discounting occurred, which consisted of five response periods that are preceded by two forced choice trials. Delays for the larger reinforcer lever were increased in ascending order (1, 3.2, 10, 32, 100 s), and proportion of choice for the larger reinforcer was recorded as the dependent variable.

For (2), we utilized publicly available RNAseq datasets from male laboratory rats and mice that were exposed to different modalities of ELA (limited nesting/bedding, maternal separation) in early postnatal life, yet all studies had prefrontal cortex (PFC) tissues microdissected and subjected to RNAseq in adulthood. GEO numbers: GSE124387, GSE153043, GSE180055, and GSE8992. We consolidated the sample information from each study and

combined count data into a single count matrix. We mapped the rat gene symbols to the mouse gene symbols. We performed principal components analysis (PCA) to identify cross-study sources of heterogeneity. Three PCs were identified and removed. We generated differentially expressed genes (DEGs), while controlling for PCs comparing ELA to control conditions (5% FDR correction), and performed weighted gene correlation network analysis (WGCNA) of these merged studies to identify transcendent PFC gene networks associated with ELA.

For (3), we examined key environmental influences on developmental precursors to substance misuse. Specifically, we conducted initial regression models of associations between the impacts of key predictors, parent income and parent education (i.e., SES), trauma, BMI, and the covariate sex on outcomes in the domains of social, novelty/impulsivity, anxiety, frustration tolerance, social, and memory deficits.

Results: For (1), resource groups are being powered in the delay discounting task. However, we observe delay-dependent decreases in the proportion of choice for the larger reinforcer in all groups (Two-way ANOVA, main effect of delay, $P < 0.05$).

For (2), we observe significant DEGs across ELA versus control conditions ($P < 0.05$). For WGCNA, we constructed 'signed' networks and identified 41 gene modules. We performed module-trait analyses. We identified five gene molecules significantly associated with ELA, but no gene modules associated with study (which indicates that study batch effect is well controlled). Modules significantly associated ($P < 0.05$) with ELA were: pink and purple (positively); lightcyan, tan, orange (negatively). By performing gene-set enrichment analyses (GSEA) on the genes of each of the five ELA-associated modules, we see the positively correlated modules are enriched for genes related to G protein activity and phosphatase activity (pink) and transporter/channel regulation (purple); whereas the negatively correlated modules are enriched for genes related to hormone receptor activity and ion channel activity (lightcyan), components of the myelin sheath (tan), and neuropeptide signaling (orange) (all at $P < 0.05$).

For (3), we see consistent main effects ($P < 0.05$) of SES on social, novelty/impulsivity and memory constructs (i.e., social responsiveness, behavioral inhibition, delay discounting, alcohol sips, picture memory), but few effects of SES on anxiety or frustration tolerance constructs. Trauma is inconsistently associated with social and novelty/impulsivity, and anxiety behaviors. BMI was associated with social and anxiety outcomes.

Conclusions: While still in process, we have resolved brain gene expression signatures associated with ELA in rodents, identified an impact of SES on adolescent phenotypes in humans, and have established an experimental context to interrogate neurocognitive performance following early life resource scarcity. Collectively, this cross-species work may aid in illuminating brain and behavioral motifs that precede SUDs.

Keywords: RNA Sequencing, Early-life stress/adversity, Neurocognitive assessment

Disclosure: Nothing to disclose.

P16. CANNABINOID RECEPTOR 1 AVAILABILITY IN INDIVIDUALS WITH CHILDHOOD TRAUMA: A POSITRON EMISSION TOMOGRAPHY STUDY

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Background: Childhood trauma is a major risk factor for many psychiatric disorders based on decades of research. However, the underlying mechanisms are still being investigated. The endocannabinoid (eCB) system is central to stress regulation, and at the same time, is being affected by chronic stress and trauma exposure, especially when they occur during childhood, by disrupting the natural course of eCB system development. Despite consistent evidence from animal studies on the impact of childhood trauma and chronic stress on the eCB system, human studies are contradictory. However, human studies mainly measure the peripheral levels of eCBs, which has multiple central and peripheral sources, and often do not correlate with central eCBs. To address this gap, we assessed the availability of brain cannabinoid receptor type 1 (CB1R) in individuals with childhood trauma (CT) compared to healthy controls without CT (HCs).

Methods: Adult men and women (age ≥ 18) with CT histories ($n = 22$) comprised a CT group, and age- and sex-matched healthy individuals without CT histories served as the HC group ($n = 22$). Inclusion in the CT group was determined based on exposure to a major traumatic event based on Criterion A of the DSM-5 criteria for PTSD, assessed by the Clinician-Administered PTSD Scale for DSM-5 (CAPS-5) or Structured Clinical Interview for DSM-5 Disorders (SCID-5), and exposure to traumatic events for the first time before the age of 18, assessed by the Trauma Questionnaire (TQ) and Childhood Trauma Questionnaire (CTQ). Cannabinoid receptor type 1 (CB1R) availability was measured using positron emission tomography (PET) imaging with the CB1R-specific radiotracer [^{11}C]OMAR. Using linear models, the effect of the group was assessed on global and trauma-relevant brain regions (amygdala, hippocampus, and frontal cortex).

Results: Lower composite CB1R availability was observed in the CT versus HC groups (difference -11.36% , $F(1,42) = 4.35$; $p = 0.04$, $d = 0.63$). In the LMM examining the pre-selected 3 PTSD-relevant ROIs, the group-by-region interactions was significant ($F(2,84) = 4.49$, $p = 0.014$) driving by lower CB1R availability among CT participants in the amygdala (-13.70% , $F(1,84) = 6.66$, $p = 0.01$, $d = 0.72$) and hippocampus (-14.50% , $F(1,84) = 6.59$, $p = 0.01$, $d = 0.78$), but not in the frontal region (-8.08% , $F(1,84) = 2.17$, $p = 0.14$, $d = 0.47$). There was no difference in CB1R availability based on meeting criteria for PTSD ($F(1,20) = 4.01$, $pSD \text{ Sidak} = 0.27$) or MDD ($F(1,20) = 4.11$, $pSD \text{ Sidak} = 0.27$). On average, CT participants reported encountering 19.0 ± 33.4 traumatic events in their lifetime, and event frequency was not associated with CB1R availability (Spearman $r = -0.05$, $pSD \text{ Sidak} = 0.84$). No differences were observed between participants who were or were not taking antidepressant medications ($F(1,20) = 2.63$, $pSD \text{ Sidak} = 0.32$).

Conclusions: This preliminary result of lower CB1R availability in adults with CT compared with HCs suggests eCB dysregulation associated with CT. Future studies should replicate and extend this finding and examine the potential effects of various trauma features on the eCB system.

Keywords: Childhood trauma, endocannabinoid system, cannabinoid receptor type 1, PET imaging

Disclosure: Nothing to disclose.

P17. SOCIOECONOMIC STATUS MODERATES THE INTERGENERATIONAL TRANSMISSION OF CHILDHOOD TRAUMA

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Background: Childhood maltreatment (CM) is strongly associated with elevated risk for adverse psychiatric and health outcomes. Evidence further suggests that these negative effects may extend across generations. However, it remains unclear how the intergenerational transmission of trauma may be modified by contextual factors, such as socioeconomic context. Parental education, a key index of family socioeconomic conditions, may play a critical role in shaping resilience versus vulnerability across generations. Biological aging is one measure that may capture the cumulative impact of stress and adversity across generations. Epigenetic age acceleration (EAA), derived from DNA methylation patterns, is one particularly useful and widely implemented method used to quantify biological aging in response to childhood maltreatment. Here, we investigated whether socioeconomic status, indexed by caregiver educational attainment level, moderates the association between parental childhood maltreatment and child epigenetic aging.

Methods: DNA methylation data were drawn from 585 saliva samples from 260 families across four independent studies of the intergenerational impacts of trauma (age range 4 months to 13 years). Epigenetic age was calculated using the PedBE epigenetic clock. Epigenetic age acceleration (EAA) was then quantified by taking the residuals from regressing epigenetic on chronological age. To test whether caregiver education moderated the association between caregiver childhood maltreatment (CM) and child EAA, we fit a linear mixed-effects model that also included covariates of age squared, sex of the child, epithelial and immune cell content, and methylation array, with a random effect of family.

Results: The effect of caregiver childhood maltreatment history on child EAA varied by caregiver education level, $\text{CM} \times \text{education}$ interaction: $\beta = 0.27$, $\text{SE} = 0.13$, 95% CI [0.03, 0.52], $p < 0.05$. In the low-education group, caregiver CM was negatively associated with child EAA ($\beta = -0.23$, $\text{SE} = 0.08$, 95% CI [-0.49, -0.07], $p < 0.005$), whereas no significant association was observed in the high-education group.

Conclusions: These findings suggest that the intergenerational biological embedding of trauma may vary by socioeconomic context. Here, we observed an association between caregiver history of childhood maltreatment and lower offspring EAA only among caregivers with lower educational attainment. These findings potentially reflect differences in biological stress response patterns among children of caregivers with lower versus higher educational attainment. Together, these results show the importance of social factors in the transmission of trauma effects across generations.

Keywords: Early-life stress/adversity, Intergenerational Transmission, Epigenetic Aging, Socioeconomic Status (SES), childhood maltreatment

Disclosure: Nothing to disclose.

P18. EARLY LIFE TRAUMA IS ASSOCIATED WITH ALTERATIONS IN BASAL MITOCHONDRIAL FUNCTION IN WOMEN WITH HIV

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Background: Early life trauma (e.g., physical abuse and neglect) are highly prevalent among women with HIV (WWH) and are risk factors for the development of brain health disorders including neurological (e.g., cognitive impairment) and mental health disorders (e.g., depression, anxiety, post-traumatic stress disorder). To date, most studies have focused on alterations in the stress response and immune systems as a function of early life trauma in people with and without HIV. An understudied mechanism in HIV that intersects with both the stress and immune systems is the

role of mitochondria. Mitochondria are significant contributors to cellular metabolism and are essential to normal neuronal function under homeostatic conditions. Outside of HIV, mitochondrial dysfunction has been linked to brain health disorders. However, it remains unknown if mitochondrial dysfunction is linked to early life trauma in people with HIV, particularly among WWH. Thus, we aimed to examine early life trauma as a contributor to mitochondrial dysfunction in WWH with a high prevalence of brain health disorders.

Methods: Sixty-four WWH completed the Childhood Trauma Questionnaire (CTQ) and a blood draw. Peripheral blood mononuclear cells (PBMCs) were isolated and used to test mitochondrial function via the Seahorse Cell Mito Stress Test. The Seahorse provides a functional assessment of mitochondria via measuring different components of the electron transport chain, and yields measures of basal respiration, proton leak, non-mitochondrial respiration, ATP-linked respiration, spare capacity, and coupling efficiency. We conducted a series of Spearman's rank correlations between the CTQ total score and basal mitochondrial respiration to understand if a history of early life stress is associated with peripheral mitochondrial function in adulthood, as well as the other aspects of mitochondrial function in our population.

Results: Women had a mean age of 54.7 (SD = 8.2), 93% were Black, 61% had a lifetime diagnosis of major depressive disorder via the SCID-V, and 64% met criteria for cognitive impairment on 2 or more domains. Among WWH, there was a positive association between the CTQ total score and basal respiration ($r = 0.327$, $p = 0.0088$), such that a higher CTQ score was associated with increased basal mitochondrial function. Increased basal mitochondrial function is concerning considering that over-active mitochondria have been shown to cause long-term cellular damage and eventual cell death.

Conclusions: Findings suggest that the link between early life trauma and brain health disorders in WWH may be partially driven by mitochondrial function. Additional work will examine mitochondrial function as a potential mediator of the link between early life trauma and brain health disorders in this population. These results underscore the importance of considering cellular bioenergetics as a biological pathway through which psychosocial adversity exerts long-term effects on health. By identifying changes in mitochondrial activity in relationship to a history of early life stress, this work highlights a novel target for intervention that could inform both preventative and therapeutic strategies for WWH who experience high trauma exposure. Future research should also investigate whether interventions that influence mitochondrial function, such as lifestyle modifications, pharmacologic approaches, or behavioral therapies, can mitigate trauma-related risk for neurocognitive impairment and psychiatric comorbidities.

Keywords: Mitochondria, HIV, women, Early life stress

Disclosure: Nothing to disclose.

P19. ADDITIVE EFFECTS OF ALCOHOL USE DISORDER AND EARLY LIFE STRESS ON NEGATIVE EMOTIONALITY

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Background: Early life stress (ELS) is a well-established risk for a wide range of psychiatric illness, including alcohol use disorder (AUD). A growing literature suggests that individuals with the same DSM-5 diagnosis differ clinically and biologically depending on their history of ELS exposure. We recently demonstrated that high-ELS exposure, defined by 4+ Adverse Childhood Experiences

(ACEs), is associated with a distinct neuroclinical profile in AUD, characterized by heightened negative emotionality. These findings highlight the importance of ELS history in shaping addiction-related phenotypes, however, replication in independent samples and extensions to case-control comparisons are needed.

The present study aimed to: (1) replicate associations between high-ELS and negative emotionality, now extending the scope to incorporate the full Addictions Neuroclinical Assessment (ANA) framework, including negative emotionality, incentive salience, executive function; and (2) extend this work by testing additive and interactive effects of AUD and ELS on clinical outcomes in a case-control design.

Methods: Adults with current AUD ($n = 114$, 57M/57F, age = 37.9 + 13.0) and matched controls without AUD ($n = 104$, 48M/56F, age = 34.5 + 13.3) completed the Adverse Childhood Experiences (ACE) Questionnaire and phenotypic battery as part of two studies. Participants were classified as having "no-ELS," (ACE = 0) "moderate-ELS," (ACE = 1, 2 or 3) or "high-ELS" (ACE = 4+). The ANA domains, incentive salience, negative emotionality, and executive function, were derived via Principal Components Analysis (PCA). Resulting domain scores were used to characterize the neuroclinical profiles of individuals with AUD. Within the AUD sample, we examined ELS group differences in ANA domains using univariate ANOVA. For case-control analyses, we re-derived PCA components excluding incentive salience, as control participants showed limited variance on all of these items. This yielded two domains, negative emotionality and executive function, that could be compared across AUD and control groups. We then conducted 2 (AUD vs. control) by 3 (no-, moderate-, high-ELS) ANOVAs to test main effects and interactions.

Results: Within the AUD group, 25% of participants were classified as no-ELS ($n = 28$), 49% as moderate-ELS ($n = 56$), and 26% as high-ELS ($n = 30$). There was a significant main effect of ELS on negative emotionality ($F = 4.6$, $p = 0.01$), such that individuals in the high-ELS group reported greater negative emotionality compared to diagnostic controls in the no-ELS ($p = 0.05$) and moderate-ELS ($p = 0.003$) groups. There were no effects of ELS group on incentive salience ($F = 0.9$, $p = 0.4$) or executive function ($F = 0.005$, $p = 1.0$) domains.

In the control group, 41% of participants were classified as no-ELS ($n = 43$), 47% as moderate-ELS ($n = 49$), and 12% as high-ELS ($n = 12$). A significant AUD group by ELS group interaction emerged for executive function ($F = 3.4$, $p = 0.03$). Specifically, the no-ELS control group demonstrated significantly better executive functioning compared to controls with moderate-ELS ($p < 0.001$) and high-ELS ($p = 0.04$), as well as compared to individuals with AUD and no-ELS ($p = 0.007$). There were no other differences in executive functioning between groups (all p 's > 0.5). There were main effects of AUD group ($F = 78.3$, $p < 0.001$) and ELS group ($F = 8.8$, $p < 0.001$) on negative emotionality. Across groups, individuals with AUD exhibited higher negative emotionality than controls, and those with high-ELS exhibited higher negative emotionality than participants with no-ELS or moderate-ELS.

Conclusions: These findings replicate prior work showing that individuals with high-ELS (4+ ACEs) and AUD exhibit greater negative emotionality compared to diagnostic controls with no- or moderate-ELS. This strengthens confidence in the robustness of the ELS and negative emotionality association in AUD. In a case-control analysis, we observed main effects of both AUD group and ELS group on negative emotionality, suggesting that each independently contributes to heightened emotional dysregulation. The combination of AUD and ELS appears additive, rather than interactive; both confer increased risk, but there were no synergistic interactions. In contrast, executive function showed an AUD group by ELS group interaction such that controls with no-ELS exhibited the strongest executive functioning. This could suggest that ELS compromises cognitive processes in the absence

of AUD, and that the effects of AUD mask these differences. Overall, findings suggest that ELS may define clinically distinct subtypes within AUD characterized by elevated negative emotionality. This aligns with the broader literature suggesting that individuals with the same DSM-5 diagnosis differ in their clinical presentation based on ELS history. Results underscore the importance of incorporating ELS history into assessment and, possibly, treatment planning for AUD.

Keywords: Early life stress, alcohol use disorder, negative emotionality, executive function

Disclosure: Nothing to disclose.

P20. GENOME-WIDE DNA METHYLATION ANALYSIS REVEALS EPIGENETIC SIGNATURES OF EARLY LIFE STRESS IN MAJOR DEPRESSIVE DISORDER PATIENTS

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Background: Major Depressive Disorder (MDD) is a pervasive condition, affecting over 20% of the global population, and is a significant burden to modern society. The etiology of MDD remains complex despite significant progress in understanding its pathology and treatment. ELS, encompassing events like abuse and neglect, is a well-documented risk factor for the development of MDD and other psychiatric disorders in later life. Individuals exposed to ELS often exhibit distinct clinical and biological features, making the investigation of the precise mechanisms by which ELS increases susceptibility to mood disorders a critical area of study. A significant dimension of this relationship involves epigenetic modifications, which are increasingly recognized as key mechanisms through which environmental exposures, such as ELS, interact with an individual's genetic predispositions to shape the depression phenotype. The present study investigated genome-wide DNA methylation profiles in individuals with MDD, stratified by ELS exposure, and included trauma-matched control groups. By comparing MDD and control participants with and without ELS, this study aimed to identify differentially methylated regions (DMRs) that are specifically associated with ELS-related depression. Additionally, we explored the biological relevance of these epigenetic modifications through functional annotation and pathway enrichment to elucidate mechanistic pathways and identify potential targets for intervention.

Methods: The participants included 90 male and female adult patients aged 18–65 who met DSM-5 diagnostic criteria for MDD or volunteer controls as determined by the MINI International Neuropsychiatric Interview. Subjects comprised of MDD with ELS ($n = 30$), MDD without ELS ($n = 30$), and non-psychiatric controls with and without ELS ($n = 30$). The UAB IRB approved the protocol, and written informed consent was obtained from all participants. Early life trauma was assessed using the Childhood Trauma Questionnaire (CTQ). Genome-wide methylation using reduced representation bisulfite sequencing. Differential methylation was evaluated by statistical tests conducted at each CpG location to determine significance, subsequently followed by the identification of differentially methylated regions (DMRs). Individual CpG sites were tested using the Wald test in Dispersion Shrinkage for Sequencing Data and Fisher's exact test in MethylKit, with raw p -values ($p < 0.05$) used for initial filtering, as is standard in DMR detection pipelines to avoid overly conservative thresholds at the single-CpG level. The omics annotations derived from the ANNOVAR framework were integrated to determine biological implications and the CNS trait association of the DMR genes. Given

the dense distribution of CpGs in and around the quantified DMR regions, localized genomic visualization models were created using the UCSC genome Browser to investigate the support of a strong DMR signal. To explore the functional relevance of genes associated with the DMRs, GENE2FUNC module in the Functional Mapping and Annotation (FUMA) platform was used.

Results: We identified 128 DMRs in MDD vs. control, 29 in MDD + ELS vs. control+ELS, and 13 in MDD non-ELS vs. control non-ELS comparisons. Notable genes exhibiting DMRs in MDD + ELS included LONRF1, MAPK8IP1, SELENOO, PDXP, and CNNM3, showing strong enrichment in brain-related tissues and regulatory elements. Functional enrichment highlighted MAPK, synaptic vesicle, and JAK-STAT pathways. In contrast, MDD non-ELS DMRs included genes such as BCL7B, BLCAP, and NNAT, which exhibited elevated expression in multiple brain regions and were enriched in Hippo, Wnt, and Ras signaling pathways and chromatin remodeling.

Conclusions: This study provides compelling evidence of distinct DNA methylation signatures associated with ELS-induced MDD and MDD without ELS exposure, highlighting the complex interplay between genetic and environmental factors in the etiology of this complex disorder. Our findings suggest that ELS may potentiate epigenetic changes in the context of MDD, leading to differential methylation patterns in genes involved in neuronal signaling, stress response, and immune function. These results contribute to our understanding of the molecular mechanisms underlying ELS-induced MDD and may have implications for the development of novel diagnostic and therapeutic strategies. Future studies integrating methylation, gene expression, and functional assays in both peripheral and brain tissues, with a focus on DMRs showing substantial effect sizes, are essential to validate their biological relevance to MDD and ELS.

Keywords: Epigenetics, Early life stress, Major Depression Disorder, DNA Methylation

Disclosure: Nothing to disclose.

P21. FOOD INSECURITY AND SUBSEQUENT EMOTION DYSREGULATION IN ADOLESCENTS: FINDINGS FROM THE ABCD STUDY

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Background: Adolescence represents a critical period for emotional development, coinciding with reduced caregiver co-regulation and increased vulnerability to mental health difficulties. Food insecurity may represent a more proximal stressor than broad socioeconomic indicators, potentially affecting adolescent development through established family stress and resource unpredictability pathways. However, research examining food insecurity's specific relationship to parent-perceived adolescent emotion dysregulation remains limited.

Methods: Using data from the Adolescent Brain Cognitive Development (ABCD) Study, we examined the associations between household food insecurity and parent-reported difficulties in emotion regulation among 2553 adolescents. Food inaccessibility was assessed at baseline using a single-item measure of food affordability over the past 12 months. Parent-reported adolescent dietary quality (Child Nutrition Assessment) and emotion regulation (Difficulties in Emotion Regulation Scale, DERS) were also collected at years 2 and 4, respectively. We examined both overall emotion regulation difficulties and specific subscale domains while controlling for demographic factors, combined household income, and BMI.

Results: Food insecurity was reported by 5.99% of caregivers. After controlling for covariates, parents in food-insecure households reported significantly higher difficulties with emotion regulation in their adolescents ($\Delta = 4.56$, 95% CI [1.82, 7.30], $p = 0.001$). Specific parent-reported difficulties emerged in emotional awareness ($\Delta = 1.10$, $p = 0.020$), catastrophic thinking during distress ($\Delta = 1.77$, $p = 0.008$), and difficulty maintaining goal-directed behavior during emotional episodes ($\Delta = 0.99$, $p = 0.003$). Adolescent dietary quality did not moderate these relationships.

Conclusions: Food insecurity is associated with parents' perceptions of greater emotion regulation difficulties in their adolescents, particularly in domains involving emotional awareness and adaptive responding during distress. However, these associations reflect correlational relationships and should be interpreted cautiously, given the 4-year interval between the food insecurity assessment and the emotion regulation outcomes, which limits causal inference. Parents experiencing food insecurity may themselves experience elevated stress, depression, or anxiety that could influence their perceptions of their adolescents' emotional functioning. Additionally, the observed differences (4.56 points) fall below established thresholds for clinical significance (≥ 9 points). While food insecurity may contribute to family dynamics that affect adolescent emotional development, these findings could equally reflect stressed parents' heightened sensitivity to their children's emotional behaviors rather than objective differences in adolescent emotion regulation capabilities. The single-item affordability measure may not capture the full complexity of food insecurity experiences. Future research should incorporate adolescent self-report measures, examine parent psychological functioning as a mediator, and use longitudinal designs with more frequent assessments to disentangle whether associations reflect actual adolescent emotion regulation differences, parental perception biases, or broader family stress processes.

Keywords: Food Insecurity, Adolescence, Emotional dysregulation, Chronic social stress, parenting distress

Disclosure: Nothing to disclose.

P22. ASSOCIATION OF ADVERSE PRENATAL EXPOSURE BURDEN WITH PERSISTENT PSYCHOPATHOLOGY AND ACCELERATED CORTICAL THINNING IN YOUTH

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Background: Adverse prenatal exposures (APEs) can disrupt critical neurodevelopmental processes during fetal development, an early sensitive period that lays the foundation for later vulnerability to psychopathology. APEs often co-occur and associate with dose-dependent effects on childhood psychopathology; however, most prior work has focused on individual APEs and their relation to specific psychiatric and brain-based outcomes. Whether exposure to multiple APEs associate with persistent clinical effects through adolescence or with alterations in cortical development remains uncertain.

Methods: Leveraging the largest U.S. population-based longitudinal and enriched prenatal exposure data from 8515 singleton children aged 9 to 15 years in the Adolescent Brain Cognitive Development (ABCD) study, we extracted cumulative burden of APEs derived from six binary prenatal exposures, 17 Child Behavior Checklist (CBCL) indices assessed annually, and 68 regions of

cortical thickness based on Desikan-Killiany atlas. Participants received up to 3 MRI scans and 5 CBCLs over a 4-year period. Generalized and linear mixed-effects models were used to examine associations of cumulative burden of APEs to (1) risk of both dimensional and clinically significant (CBCL total ≥ 60) psychopathology, (2) cortical thickness, and (3) age-by-APE interactions on clinical and neurodevelopmental trajectories, after controlling for biological (age, age², sex, pubertal stage), design-related (subject ID, family ID, study site), technical (scanner, intracranial volume, surface hole number) and socioeconomic factors. Findings were validated within 414 non-adopted sibling pairs with discordant APEs, accounting for unmeasured genetic and other familial confounders. False discovery rate (FDR) was used to control for multiple comparisons.

Results: Of the 8,515 singleton children with complete prenatal exposures (47.6% females), 78% were exposed to at least one APE. Multiple APEs were persistently associated with increased odds of clinically significant CBCL total problem scores ($\chi^2 = 69.72$; $PFDR < 0.001$). Specifically, compared with unexposed children, sequentially stronger associations were observed for exposure to one APE (OR, 2.01; 95% CI, 1.28–3.16; $PFDR = 0.0065$), two APEs (OR, 3.82; 95% CI, 2.39–6.11; $PFDR = 9.12 \times 10^{-8}$), and three or more APEs (OR, 6.75; 95% CI, 4.14–11.02; $PFDR = 1.31 \times 10^{-13}$), demonstrating a dose effect. Associations of APEs with attention-deficit/hyperactivity disorder symptoms attenuated over time (interaction: $F = 13.51$; corrected $P = 7.13 \times 10^{-8}$), while those with depressive symptoms during adolescence potentiated (interaction: $F = 5.82$; corrected $P = 0.0019$). Increased APE burden (0 to 3+) associated with dose-dependent acceleration in age-related cortical thinning in 36 of 68 cortical regions (interactions: $F = 3.26$ –8.89; corrected P 's = 0.039 – 4.86×10^{-4}), indicating a cumulative impact of prenatal exposures on cortical maturation. Importantly, the primary findings were validated in 414 sibling pairs with discordant APEs. Despite the smaller number of participants, siblings with higher exposure demonstrated persistently higher CBCL total problems ($T = 2.25$; uncorrected $P = 0.025$) and accelerated cortical thinning (interactions: $T = -3.00$ – -2.10 ; uncorrected P 's < 0.05) in 5 of the 36 regions implicated in the larger sample.

Conclusions: To our knowledge, our study provides the first evidence that exposure to multiple APEs associates with persistent, dose-dependent increases in the risk of clinically significant psychopathology and acceleration in age-associated cortical thinning during adolescence. While highlighting the importance of fetal programming to mental health across life course, these results provide clinically meaningful risk stratification for mental illness risk in adolescence, and underscore the need for tailored interventions to reduce multiple gestational adverse exposures in high-risk families.

Keywords: Adverse prenatal exposure, Brain development, Cortical thickness, Psychopathology, Adolescence

Disclosure: Nothing to disclose.

P23. RESILIENCY TO ANXIETY AND SUBSTANCE USE IN THE WAKE OF CHILDHOOD TRAUMA: THE ROLE OF REWARD PROCESSING AND SELF-COMPASSION

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Background: Childhood maltreatment has been associated with the development of anxiety and increased risk of substance use and addiction, but it is not completely understood why some individuals exhibit greater susceptibility, while others are resilient.

Since the hippocampus is critical for understanding stress susceptibility and the reward system has been proposed as a biologically plausible mechanism for stress reduction and resilience, we hypothesized that resilience to childhood maltreatment and development of anxiety will be associated with altered hippocampal connectivity during reward processing, and that this resilience-related hippocampal connectivity will predict substance use. Further, we hypothesized that self-compassion, an important resilience factor, would mediate the relationships between the hippocampal connectivity and resilience.

Methods: We conducted a neuroimaging follow-up of the European Longitudinal Study of Pregnancy and Childhood (ELSPAC) prenatal birth cohort in young adulthood ($n = 114$, 45.6% men, 28–30 years of age), allowing us to detect small effects ($r = 0.26$ or higher) in the whole sample and medium effects ($r = 0.37$ or higher) in the sex-specific analyses with a power of 80% and alpha of 0.05. Similarly to Ioannidis et al (2020), we took an outcome-oriented approach to quantify resilience and inferred the level of resilient functioning from the residuals of the relationship between childhood maltreatment, assessed using the Childhood Trauma Questionnaire (CTQ; Bernstein et al. 2003) at the age of 18, and anxiety trait, assessed using Spielberger's Trait Anxiety Inventory (STAI-T; Spielberger et al. 1983) at the age of 28–30. Thus, the residual scores reflect the extent to which an individual functioned better than expected or worse than expected given their history of childhood maltreatment.

Structural and functional magnetic resonance imaging (MRI, fMRI) was acquired using a 3 T Siemens Prisma MRI Scanner at the age of 28–30. Reward processing in the young adult offspring was tested using a Monetary Incentive Delay (MID) fMRI task (Knutson et al. 2000). The pre-processing of the fMRI data was conducted as in our prior work (Mareckova et al. 2024). Next, we used the Psychophysiological Interaction (PPI) analysis (Friston et al. 1997) to study how the reward task conditions moderate functional connectivity of the hippocampus. A general linear model tested the effects of Resilience, Sex, and their interaction on PPI connectivity from the left and right hippocampus. Results were considered significant if they survived an initial voxelwise threshold $p < 0.001$ uncorrected as well as cluster-level family-wise-error (FWE) rate corrected $p < 0.05$. Next, a linear regression evaluated the relationship between resilience and self-compassion, assessed using the Self-Compassion Scale Short Form (SCS; Raes et al., 2011), and the relationship between resilience-related hippocampal connectivity and substance use. The role of Sex and potential interactions with Sex were evaluated. Finally, mediation analysis tested the role of self-compassion in the relationship between hippocampal connectivity during reward and resilience to childhood maltreatment and the subsequent development of anxiety.

Results: Those more resilient to childhood maltreatment and development of anxiety were more self-compassionate ($R^2 = 0.45$, $p < 0.0001$) and had a lower difference in hippocampal connectivity between positive and negative reward feedback, particularly in 7 regions responsible for emotion regulation and self-reassuring thoughts, attention and error monitoring, reward-related decision making, and memory for rewards (FWE $p < 0.05$). Further, women (but not men) with a greater difference in hippocampal - supramarginal gyrus connectivity during reward processing exhibited greater substance use ($R^2 = 0.08$ – 0.11 , $p < 0.027$). Moreover, self-compassion mediated the relationship between the hippocampal connectivity during reward processing and resilience to childhood maltreatment and the development of anxiety. The mediation was significant in 6 out of the 7 resilience-related clusters (right temporal pole: $ab = -0.07$, $SE = 0.03$, 95% CI $[-0.13; -0.01]$; anterior cingulate: $ab = -0.08$, $SE = 0.03$, 95% CI $[-0.14; -0.03]$; right hippocampus: $ab = -0.07$, $SE = 0.03$, 95% CI $[-0.14; -0.02]$; left supramarginal: $ab = -0.06$, $SE = 0.03$, 95% CI $[-0.11; -0.01]$; right supramarginal: $ab = -0.06$, $SE = 0.03$, 95% CI $[-0.12; -0.01]$; left rectus/OFC ($ab = -0.07$, $SE = 0.03$, 95% CI $[-0.13; -0.01]$).

Conclusions: Our findings reveal that resilience to childhood maltreatment and the associated development of anxiety in young adulthood is characterized by lower differences in hippocampal connectivity during processing of positive vs. negative reward feedback, suggesting that better resilience might be associated with lower reward dependence. Consistently, we showed that in women, a greater difference in hippocampal connectivity with the supramarginal gyrus during reward processing was associated with greater substance use. Since self-compassion mediated the relationship between the hippocampal connectivity during reward feedback and resilience, our findings also reveal that treating oneself with kindness, understanding, and acceptance, especially during stress and failure, might protect high-risk individuals against the development of anxiety and addiction.

Keywords: Resilience to childhood maltreatment and anxiety, reward processing, hippocampal connectivity, self-compassion, substance use

Disclosure: Nothing to disclose.

P24. INTERSECTING EPIGENETIC MECHANISMS OF EARLY LIFE ADVERSITY AND PHYSICAL EXERCISE REVEAL NEW TARGETS FOR INTERVENTION

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Background: Early-life adversity (ELA) can predispose to long-term cognitive impairments, yet effective treatments to buffer against these consequences do not exist. Our previously published data demonstrate that voluntary wheel running specifically during the juvenile developmental stage in mice (Juv EX) results in lasting improvements in hippocampal memory and synaptic plasticity. We also found that Juv EX intervention can mitigate memory deficits in middle-aged ELA mice, and characterized the hippocampal gene expression profile underlying Juv EX intervention in preserving cognitive functions. Employing the Emx1-NuTRAP transgenic mouse line, we performed transcriptomic and epigenomic sequencing in hippocampal excitatory neurons and identified the lysine demethylase enzyme KDM7a as a candidate epigenetic mediator of Juv EX effects. Here, we test the hypothesis that Juv EX mitigates cognitive impairments after ELA by promoting a transcriptionally permissive epigenetic state, and expression of memory-related genes, via the activity of KDM7a.

Methods: Emx1-NuTRAP mice ($n = 6$ – 10 mice/sex/group) were used to populate four experimental groups: Control, ELA, Juv EX, and ELA+Juv EX intervention. ELA consisted of limiting bedding and nesting material in the cage during postnatal days (P) 2–9. On P21, Juv EX mice were pair-housed in running wheel-equipped cages and running distances were monitored until P42. In adulthood (10–14 months), mice underwent a battery behavior tasks. Mice were sacrificed at P21, P42, or adulthood, and bilateral dorsal hippocampi were collected for molecular studies. DESeq2, SEACR, and Panther GO (sequencing studies) and One- and Two-way ANOVA with post-hoc tests (mass spec, western blot and RT-qPCR data) were used. To test KDM7a regulation of H3K27 methylation and memory-associated gene expression, siRNA was used in vitro to evaluate methylation status and gene expression.

Results: RNA-seq demonstrates enhanced specific memory and plasticity genes after Juv EX, and ELA+Juv EX intervention ($lfc < 0.05$). CUT and RUN-seq heat maps demonstrate greater H3K27me1 peak distribution across the genome following Juv EX alone and after ELA. Mass spectrometry revealed a distinct

epigenetic pattern of histone modifications after ELA, that was altered after Juv EX intervention. KDM7a gene was significantly increased in males, but not females, after Juv EX and EX intervention following ELA (one-way ANOVA, $p < 0.05$ using Bonferroni's post-hoc test). There was a higher abundance of H3K27me2 after ELA exposure, whereas Juv EX groups (with or without prior ELA exposure) had greater abundance of H3K27me1. KDM7a expression was blunted and memory associated gene expression was altered by in vitro siRNA experiments.

Conclusions: These data reveal distinct alterations to the epigenetic landscape after ELA, Juv EX, and Juv EX as an intervention after ELA. We also implicate an epigenetic mechanism by which Juv EX modulates hippocampal memory in adulthood, potentially through KDM7A demethylase activity at H3K27, leading to gene expression profiles that support hippocampal memory.

Keywords: Epigenetic Modification, early-life adversity, Hippocampal Function, exercise, ChIP-sequencing

Disclosure: Nothing to disclose.

P25. VENTRAL HIPPOCAMPAL CA1 ACTIVITY ASSOCIATED WITH REDUCED BEHAVIORAL DISCRIMINATION AFTER EARLY LIFE ADVERSITY IN MICE

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Background: Early life adversity (ELA) increases vulnerability to psychiatric illnesses throughout the lifespan, and understanding how ELA affects the neural circuits controlling motivated behaviors, even in adulthood, could lead to strategies for enhancing resilience. In mouse models for ELA, such as the limited bedding and nesting model, adult mice exhibit changes in motivated behaviors months after the postnatal manipulation. In this study, we investigate the underlying circuit of these behavioral changes, focusing on the CA1 region of the ventral hippocampus. The ventral hippocampus has direct connections to both fear and reward circuits, and is analogous to the anterior hippocampus in humans—a region that frequently exhibits abnormalities in neuropsychiatric patients and survivors of ELA. We recorded calcium activity from the ventral CA1 (vCA1) during tasks with both positive and negative stimuli and evaluated behavioral discrimination.

Methods: ELA is induced in c57BL6 pups using the limited bedding and nesting paradigm between p4 and p11. In adulthood (12–20 weeks old), male and female mice are injected with AAV-encoded GCaMP6f under a calcium/calmodulin-dependent protein kinase II (CaMKII) promoter to target vCA1 pyramidal cells. A gradient-index (GRIN) lens is chronically implanted in the vCA1, and then calcium activity is recorded during both a sucrose gustometer task and a fear context discrimination task. The gustometer apparatus administers lick-activated droplets of water or 4 different concentrations of sucrose, presented in pseudo-random order, and records consummatory and anticipatory licks. Aerial videos are simultaneously recorded with calcium activity in a familiar holding cage, the gustometer arena, and in three different fear discrimination contexts (shock context, similar, and novel) for behavioral analyses. Calcium activity is preprocessed, motion corrected, and segmented for individual cell activity. $N = 9$ –13/sex/group for gustometer behavior, 3–5/sex/group with calcium data. $N = 3$ –6/sex/group for fear context discrimination,

5–7/group with calcium data. The average number of active vCA1 cells per mouse was ~ 70 .

Results: In the gustometer task, post-consummatory licking increases with increasing concentrations of sucrose, and the rate of subject-initiated trials remains steady across 30 minutes of testing. However, vCA1 calcium activity gradually increases over the course of the session, and within trials, calcium activity was lower during the reward epochs. Increased licking with higher sucrose concentrations is significantly blunted in male but not female ELA mice. In both sexes and across task epochs, there is higher vCA1 calcium activity in ELA mice, though this difference is more pronounced in males. Across groups, successful fear context discrimination is accompanied by a change in calcium activity in the neutral context, with no activity change in the shock-associated context. ELA mice show impaired contextual fear learning, more evident in females than males. Both sexes showed impaired fear context discrimination after ELA. ELA mice also exhibit alterations in vCA1 calcium activity during various phases of the fear discrimination task. $p < 0.05$ for significance in all analyses.

Conclusions: Our findings demonstrate that ELA alters processing of both positive and negative valence stimuli in the vCA1, with some observed sex differences. We also present novel properties of vCA1 response during a reward-seeking task. Given the widespread outputs of the vCA1, including to networks mediating fear, reward, motivation, and cognition, these changes likely affect the broader network of emotion-related processing.

Keywords: ventral hippocampus, pattern separation, Reward, contextual fear, in vivo calcium imaging

Disclosure: Nothing to disclose.

P26. SEX-SPECIFIC NEUROVASCULAR AND ASTROCYTE ALTERATIONS IN THE MEDIAL PREFRONTAL CORTEX FOLLOWING EARLY-LIFE ADVERSITY

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Background: The brain vasculature function is essential for brain homeostasis, and its disruption is increasingly recognized as a critical contributor to cognitive decline and dementia. The neurovascular unit (NVU) is composed of endothelial cells, astrocytes, and pericytes. Astrocytes contact blood vessels through astrocyte endfeet that express Aquaporin-4 (AQP4). Chronic stress in adulthood can impact the NVU. However, few studies have examined whether early adversity affects the NVU, which is surprising given that the NVU develops well into the postnatal period. Our laboratory found that limited bedding and nesting (LBN), an early life resource scarcity manipulation, alters vascular-associated gene expression in the medial prefrontal cortex (mPFC) of adult female but not male rats. Here we tested whether LBN induces persistent, sex-specific NVU remodeling.

Methods: Long-Evans rats were placed under LBN or control conditions from postnatal day (PND) 2–10. In LBN, dams and pups are separated from their bedding and given only a single paper towel as nesting material. Control animals had access to adequate bedding and nesting materials and enrichment. Neurovascular morphology was assessed in the adult mPFC using CD31 and AQP4 immunohistochemistry and quantified in Imaris. Astrocytic morphology was assessed by AAV-GfaABC1D-Lck-GFP tracing, a virus with a cell surface label that allows for thorough astrocyte reconstruction, and number and reactivity with GFAP labeling. Based on our sequencing data we a priori predicted that there would be an effect of LBN on the NVU endpoints in females but

not males so we ran planned comparisons based on sex. Sholl branching by mixed factors (with repeated-measures) ANOVA.

Results: We used CD31+ labeling to assess whether LBN affected the volume of endothelial cells in the mPFC of adult male and female rats. Planned comparisons test revealed that female LBN rats had significantly reduced CD31+ volume compared with female controls ($p = 0.0226$, $\eta^2p = 0.22$, $d = 1.42$), whereas no difference was observed between male groups ($p = 0.689$, $\eta^2p = 0.01$, $d = 0.23$; $n = 6$ – 7 /group, 8 – 12 images/animal).

We next analyzed vessel surface number as a proxy for the number of blood vessels in the mPFC. There were no significant interaction or treatment effect, but there was a main effect of sex ($F(1,21) = 8.462$, $p = 0.008$). Planned comparisons indicated that LBN females had significantly reduced surface number compared with LBN males ($p = 0.0031$, $\eta^2p = 0.42$, $d = 2.15$), whereas no differences were observed in the other group comparisons.

Ongoing studies are analyzing filament length (currently only $n = 2$ – 4 /group).). However, our preliminary data reveal a trend for a main effect of LBN ($F(1,7) = 3.769$, $p = 0.093$, $\eta^2p = 0.35$). Planned comparisons indicated a trend toward shorter filament length in LBN females compared with controls ($p = 0.065$, $\eta^2p = 0.41$, $d = 1.7$), but no difference was observed in males ($p = 0.466$, $\eta^2p = 0.07$, $d = 0.7$).

Our filament branching analysis is also preliminary but planned comparisons showed reduced branching in LBN females compared with controls (mean diff = 8.736, 95% CI [0.907–16.56], $p = 0.034$, $\eta^2p = 0.55$, $d = 2.2$), whereas no difference was observed in males ($p = 0.866$, $\eta^2p = 0.01$, $d = 0.18$). Together, these data reveal that LBN reduces vessel coverage in the mPFC of adult female but not male rats. Preliminary data indicate that this reduction in coverage is likely due to shorter and less complex vessels in LBN females.

AQP4 endfeet volume revealed no significant main effects of sex ($F(1,19) = 0.565$, $p = 0.461$), condition ($F(1,19) = 1.224$, $p = 0.282$), or sex $\times$ condition interaction ($F(1,19) = 0.740$, $p = 0.401$). Planned comparisons confirmed no differences between LBN and control groups in either females ($p = 0.190$, $\eta^2p = 0.09$, $d = 0.83$) or males ($p = 0.860$, $\eta^2p = 0.002$, $d = 0.08$).

Sholl analysis of astrocytes revealed increased dendritic branching in both sexes. Females showed a significant main effect of group ($F(1,31) = 7.377$, $p = 0.011$) and a radius $\times$ group interaction ($F(77,2387) = 4.454$, $p = 0.014$), with LBN females displaying greater branching than controls (mean diff = 4.90, $d = 0.95$). Males also exhibited a significant group effect ($F(1,38) = 6.131$, $p = 0.018$) and interaction ($F(77,2926) = 2.98$, $p = 0.027$), with LBN males showing increased branching compared with controls (mean diff = 2.54, $d = 0.78$).

Conclusions: Our findings suggests LBN induces persistent NVU remodeling in the mPFC, with female-biased vessels vulnerability. These findings resemble features of accelerated vascular aging and highlight sex as a determinant of NVU vulnerability to early adversity. This is consistent with prior findings showing females have higher vulnerability for vascular aging and dementia. Further investigation is needed to evaluate the functional consequences of these morphological modifications. Endothelial–astrocytic interactions may represent a key pathway linking stress exposure to long-term brain health in a sex-specific way.

Keywords: Early life adversity, Neurovasculature, Sex-specific effects

Disclosure: Nothing to disclose.

P27. SEX-SPECIFIC CIRCUIT AND BEHAVIORAL ADAPTATIONS TO ACUTE AND ADOLESCENT STRESS EXPOSURE

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Background: The dorsal raphe nucleus (DRN) serotonin (5-HT) system is critical for emotional and social regulation. Stress, especially during early life, can produce lasting changes in socioemotional behavior and neurocircuitry. Adolescence is a sensitive period for stress-related plasticity, making the 5-HT system vulnerable to long-term effects. The lateral hypothalamus (LH), a key modulator of sensorimotor and stress signals, sends dense input to the DRN. However, how acute stress alters LH–DRN circuits, how early trauma shapes later stress responses, and how these effects vary by sex remain unclear.

Methods: Adult male and female C57BL/6 mice were exposed to acute footshock stress (10x, 0.5 mA, 2 s). To monitor LH–DRN activity using fiber photometry, mice received AAV-hSynapsin1-axon-jGCaMP8f-P2A-mRuby3 in the LH and an optical fiber over the DRN (stress/control: $n = 5/6$ females, $3/3$ males). Post-stress, mice underwent open field and social interaction tests. To assess LH-driven modulation of DRN physiology, AAV-Syn-ChR2(H134R)-GFP was injected into the LH, and whole-cell recordings were performed on DRN slices 1 h after footshock (16/3 cells/mice) or control exposure (14/3 cells/mice). In a separate cohort, a chronic unpredictable stress (CUS) paradigm was applied during adolescence (PND 28–40), and socioemotional behavior was tested in adolescence and adulthood (7 females, 3–5 males). For connectivity analysis, brains were collected 90 min after footshock for cFos quantification (12–13 females, 12–13 males).

Results: Acute footshock stress activated the LH–DRN pathway in both sexes (baseline vs. footshock: $p = 0.005$). In the open field, stressed females spent more time in the aversive center (stress vs. control: $p = 0.0126$) and showed reduced LH–DRN activity ($p = 0.012$). Acute stress reduced social sniffing in both sexes ($p = 0.022$) with reduced LH–DRN activity during social contact ($p = 0.062$). In male DRN slices, acute stress depolarized 5-HT neurons ($p = 0.012$) and increased their firing in response to current ($p < 0.001$). Optogenetic stimulation of LH terminals altered 5-HT neuron responses in stressed vs. control mice (decreased firing: $p < 0.0001$; increased firing: $p = 0.074$), suggesting stress modifies LH input. Social preference was reduced after adolescent CUS in males but increased in females (group $\times$ sex trend: $p = 0.086$). CUS also elevated passive coping in males ($p = 0.016$), with no change in females ($p = 0.33$). After acute stress, CUS mice showed reduced LH–DRN connectivity, suggesting long-term circuit disruption. Ongoing studies aim to determine whether targeting the DRN can reverse these effects.

Conclusions: Our findings show that acute footshock disrupts LH-mediated regulation of DRN activity, while adolescent chronic stress increases adult stress susceptibility by altering emotional circuit connectivity. We also identify sex-specific mechanisms in acute stress responses, underscoring the importance of developmental timing and biological sex in understanding stress-related psychiatric disorders.

Keywords: Serotonin, Dorsal raphe nucleus, Adolescence, Early life stress (ELS), Lateral hypothalamus

Disclosure: Nothing to disclose.

P28. CHRONIC ADOLESCENT STRESS AND MORPHINE DEPENDENCE INDUCE SEX-SPECIFIC ALTERATIONS IN T CELL PROFILE

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Background: Opioid use disorder (OUD) is a debilitating condition characterized by repeated cycles of craving, excessive use, and

withdrawal. Stress exposure, particularly during development, is a prominent risk factor in developing OUD. One of the mechanisms mediating stress-induced increases in OUD risk may lie within the immune system. Individuals with a history of traumatic stress exhibit decreased regulatory T cell (Treg) levels. Tregs are critical for immunosuppression, and insufficient levels can induce immune dysregulation and even autoimmune disease. Furthermore, morphine withdrawal has also been shown to induce Treg dysfunction. The aims of this study were to characterize the effects of chronic adolescent stress exposure and morphine dependence on circulating T cell profile.

Methods: Male and female Wistar rats were exposed to chronic adolescent stress (CAS) or control conditions and then chronic morphine or saline exposure began in adulthood. Blood samples were collected prior to stress, after chronic stress, prior to morphine initiation, after a single dose, after dependence, and during spontaneous withdrawal. Samples were processed for flow cytometry and run on Cytex Aurora 5-Laser Spectral Analyzer. T cell profile data were analyzed using two-way ANOVAs for the timepoints before drug exposure with the between-subjects factors of sex and CAS. Three-way ANOVAs were conducted for morphine dependence and withdrawal timepoints with the between-subjects factors of sex, CAS, and morphine. When appropriate, post-hoc analyses were conducted using Bonferroni correction. Cohen's d and partial eta squared (η^2) were calculated for effect size with t-test and ANOVA analyses, respectively.

Results: Significant interactions between sex and CAS ($F_{1,53} = 5.98$, $p = 0.02$, $\eta^2 = 0.115$), sex and morphine ($F_{1,53} = 6.71$, $p = 0.01$, $\eta^2 = 0.127$), and sex, CAS, and morphine ($F_{1,53} = 7.25$, $p = 0.01$, $\eta^2 = 0.136$) were observed for circulating Tregs 2hr following the first morphine treatment. Post-hoc analyses demonstrated a CAS-induced decrease in Treg levels in control males and females ($p < 0.001$) and a morphine dependence-induced decrease in Treg levels for subjects of both sexes without a stress history ($p < 0.001$). A sustained CAS-induced decrease in Treg levels in males ($p < 0.001$) was also observed, regardless of morphine treatment. No significant differences in Th17:Treg ratio were observed at this timepoint. After chronic morphine treatment and induction of dependence, main effects of sex, CAS, and morphine were observed for circulating Treg levels, with males having higher overall Treg levels compared to females ($F_{1,52} = 11.40$, $p < 0.01$, $\eta^2 = 0.093$), and CAS ($F_{1,52} = 14.47$, $p < 0.001$, $\eta^2 = 0.243$), and morphine ($F_{1,52} = 10.54$, $p < 0.01$, $\eta^2 = 0.190$) leading to overall decreases in Treg levels. Significant interactions between sex and CAS ($F_{1,52} = 14.39$, $p < 0.001$, $\eta^2 = 0.242$) and CAS and morphine ($F_{1,52} = 4.82$, $p = 0.03$, $\eta^2 = 0.097$) were also observed. Post-hoc analyses showed a sustained (in males) and delayed (in females) CAS-induced decrease in Treg levels ($p < 0.001$) and a morphine dependence-induced decrease in Treg levels for subjects of both sexes with no history of CAS ($p < 0.001$). A sustained CAS-induced decrease in Treg levels in males ($p < 0.001$) was also observed, regardless of morphine treatment. Analyses of Th17:Treg ratios showed a main effect of CAS ($F_{1,49} = 5.79$, $p = 0.02$, $\eta^2 = 0.121$) and a significant interaction between sex, CAS, and morphine ($F_{1,49} = 8.57$, $p < 0.01$, $\eta^2 = 0.169$). Post-hoc analysis demonstrated that CAS-VEH males had significantly higher Th17:Treg ratios than NS-VEH males ($p = 0.01$), and that CAS-MOR females had significantly higher Th17:Treg ratios than CAS-VEH females ($p = 0.02$) and NS-MOR females ($p = 0.01$). Analyses of Th17:Treg ratios showed main effect of CAS ($F_{1,49} = 5.79$, $p = 0.02$, $\eta^2 = 0.121$) and a significant interaction between sex, CAS, and morphine ($F_{1,49} = 8.57$, $p < 0.01$, $\eta^2 = 0.169$). Post-hoc analysis demonstrated that males with a history of CAS had significantly higher Th17:Treg ratios than control males ($p = 0.01$), and that females with a history of stress and morphine exposure had significantly higher Th17:Treg ratios than both females with a history of only stress ($p = 0.02$) and females with a history of only morphine ($p = 0.01$).

Conclusions: Collectively, these findings demonstrate a prolonged impact of adolescent stress exposure on immunophenotype and indicate that these changes interact with morphine-induced changes in immunophenotype to produce a proinflammatory signature. Given the known interactions between the brain and the immune system, understanding the extent to which stress and drug exposure alter immunophenotype will provide critical insight to the prolonged effects of both adolescent stress and opioid exposure.

Keywords: adolescent stress, Early-life stress/adversity, immune phenotype, opioid, sex difference

Disclosure: Nothing to disclose.

P29. ADOLESCENCE AT THE CROSSROADS OF PRENATAL STRESS VULNERABILITY: NEUROBIOLOGICAL MECHANISMS AND EARLY PROTECTIVE EFFECTS OF A KETOGENIC DIET

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Background: Exposure to early-life stress (ELS) is a well-established risk factor for the development of psychiatric disorders such as depression. These adverse experiences can trigger persistent behavioural and neurobiological alterations that often emerge in adolescence. Given these long-term consequences, there is growing interest in identifying therapeutic strategies, including dietary interventions, which may mitigate or even prevent the enduring effects of ELS. The ketogenic diet (KD), a high-fat, low-carbohydrate regimen that shifts metabolism toward ketone utilization, has gained attention for its neuroprotective and anti-inflammatory properties. Emerging evidence suggests that KD may modulate different systems that have been associated with the development of mental disorders. Based on this, we investigated whether KD could attenuate the behavioral and molecular alterations induced by prenatal stress (PNS) in adolescent rats.

Methods: Pregnant Sprague Dawley rats were subjected to PNS through three daily 45-minute restraint sessions from gestational day 14 until delivery, while control dams remained unstressed. Offspring were weaned at postnatal day 21 (P21) and assigned to either a control diet (CD) or KD for four weeks. During adolescence, the behavior of male and female offspring was assessed across psychopathological domains, including sociability and anhedonia, which are common to several mental disorders. Following sacrifice, candidate gene analyses were conducted in different brain regions to explore mechanisms potentially underlying the behavioral phenotypes in PNS-exposed animals. Statistical analyses were performed using two-way ANOVA with diet (CD or KD) and stress (CT or PNS), followed by Tukey's post hoc test where appropriate. A $p < 0.05$ was considered statistically significant.

Results: Consistent with our prior findings, PNS exposure induced emotional dysregulation in both male and female adolescent offspring, characterized by a significant impairment of social interaction. These deficits were ameliorated in PNS animals maintained on KD from weaning, suggesting a preventive effect on stress-induced sociability impairments. While PNS did not affect grooming behaviour, KD increased total grooming time, suggesting improved self-care. Notably, vulnerability to PNS - defined by behavioral impairments - was observed in approximately 50% of CD-fed animals, whereas KD significantly reduced this proportion to 22% in males and 12% in females, highlighting its protective role against stress-induced emotional dysfunction. At the molecular level, KD modulated

inflammatory and redox mechanisms in the prefrontal cortex. Specifically, KD reduced complement component C4 expression in both sexes, while C3 downregulation and GFAP suppression were restricted to males, indicating broader anti-inflammatory effects in males. Conversely, KD enhanced antioxidant responses selectively in females, upregulating Nrf2 and its target gene Gclc1, particularly in PNS-exposed females. This was accompanied by normalization of KEAP1 levels, suggesting restoration of redox homeostasis.

Conclusions: Collectively, these findings highlight the therapeutic potential of KD in preventing behavioural dysfunctions linked to early life stress and underscore the importance of considering sex as a biological variable. KD appears to exert sexually dimorphic effects: males primarily benefit from anti-inflammatory mechanisms, while females exhibit enhanced antioxidant responses. These insights support the development of personalized dietary strategies for the prevention and treatment of stress-related psychopathology.

Keywords: Early life stress (ELS), sociability, inflammation, Adolescence- Critical Period, redox dysregulation

Disclosure: Otsuka, Speakers Bureau, Self, Sumitomo Pharma, Board Member, Self, Sumitomo Pharma, Grant, Self, Sumitomo Pharma, Speakers Bureau, Self, Exeltis, Consultant, Self, Lundbeck, Speakers Bureau, Self.

P30. ANTIPSYCHOTIC POLYPHARMACY AMONG PATIENTS WITH VIOLENT BEHAVIORS IN THE FORENSIC PSYCHIATRY SYSTEM IN ONTARIO

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Background: Effective control of psychotic symptoms is crucial in forensic psychiatric settings to enhance recovery and manage different risk concerns. While antipsychotic polypharmacy is commonly employed to ensure symptom control and risk mitigation, there is limited research on its burden and relationship with violent and aggressive behaviors in forensic psychiatric populations. This study aims to examine the prevalence of antipsychotic polypharmacy among forensic psychiatry patients in Ontario and assess its relationship with recent incidents of aggression and violence.

Methods: We conducted a retrospective analysis using a database prepared from data captured in annual reports submitted to the Ontario Review Board by 12 forensic programs in Canada. We utilized statistical analyses based on negative binomial regression to evaluate associations between polypharmacy and incidences of violence.

Results: Among 1104 patients in the databases, 37.8% (n = 417) were on antipsychotic polypharmacy. The most frequent combination was with Olanzapine or Quetiapine. Significant associations were observed between antipsychotic polypharmacy and patients with increased incidences of physical violence to objects, self-harm, and verbal aggression.

Conclusions: Antipsychotic polypharmacy is prevalent among forensic psychiatry patients, especially among those with violent and aggressive behaviors. These findings underscore the need for rational and safe prescribing practices, and further research is needed to inform clinical guidelines to support patient safety and ensure effective treatment.

Keywords: Antipsychotic Polypharmacy, Forensic psychiatry, violence, Self-harm

Disclosure: Nothing to disclose.

P31. IDENTIFYING THE AGGRESSION IMPULSIVE/REACTIVE (AIR) PROFILE IN YOUTH WITH BEHAVIORAL CHALLENGES: LATENT PROFILE ANALYSIS IN REAL-WORLD CLINICAL DATA

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Background: Aggression in children and adolescents remains one of the leading causes for referral to mental health services, yet its nosology is poorly articulated. Prior secondary analyses of large outpatient research datasets identified a distinct Aggression Impulsive/Reactive (AIR) profile, characterized by heightened impulsive/reactive aggression and hyperactivity/impulsivity, with variable co-occurring mood symptoms. The present study aimed to test whether similar profiles are observed in a diverse, real-world clinical cohort spanning outpatient, inpatient, and emergency department settings, and to examine clinical correlates including comorbidity, developmental course, and adverse outcomes.

Methods: Medical records of patients aged 3–17 years presenting for psychiatric care over a 6-month interval were reviewed. Parents completed dimensional measures assessing aggression, mania, depression, hyperactivity/impulsivity, rule-breaking, and self-harm, drawn from validated scales including the R-MOAS, RAGA-16, SNAP-IV, and PGSI short forms. Latent profile analysis (LPA) was conducted with these six domains, mirroring prior analytic strategies. Profile membership was compared on demographics, diagnoses, medication use, history of abuse/neglect, and clinical outcomes including seclusion/restraint, PRN medications, and readmission.

Results: A total of 430 youth (M age = 12.5 years, 53% female) were included. LPA supported a four-profile solution consistent with prior findings. (1) High AIR + hyperactivity/impulsivity (n = 83): predominantly younger boys (M = 9.8 years), early onset of aggression (M = 5.1 years), high ADHD prevalence (78%), low mood disorder rates, and lowest likelihood of abuse history; least frequent adverse outcomes. (2) High AIR + mania/mood (n = 54): elevated mania alongside AIR, depression, and rule-breaking; included 7% with bipolar spectrum diagnoses, high rates of abuse (54%), and frequent DMDD diagnoses. (3) High AIR + mood/self-harm (n = 56): elevated self-harm, depression, and anxiety, enriched for females and ED/inpatient presentations (71%), with 61% reporting abuse history; greater risk of adverse outcomes. (4) Moderate overall symptoms (n = 237): older adolescents, majority female, high anxiety and depression prevalence, moderate AIR, and intermediate clinical risk. Across the sample, aggressive outbursts showed a bimodal onset distribution (peaks near ages 3 and 11), with boys more likely to exhibit earlier onset.

Conclusions: This study replicates and extends prior research by demonstrating the presence of distinct AIR profiles in a treatment-seeking clinical sample. The predominantly AIR + hyperactivity group emerges as a robust phenotype, distinct from mood-related profiles by age, sex distribution, onset, comorbidity, and abuse exposure. The replication across differing samples, measures, and clinical settings strengthens the construct validity of AIR as a transdiagnostic dimension. These findings underscore the need to articulate diagnostic criteria for impulsive/reactive aggression, refine its relationship to disorders such as ADHD, DMDD, and mood disorders, and explore prognostic implications including risk for adverse clinical outcomes. Future work should pursue longitudinal validation, links to neurobiological mechanisms, and development of targeted interventions to improve care for youth with AIR.

Keywords: aggression, latent profile analysis, disruptive behavior disorders

Disclosure: Joe Startup Technologies, Stock/Equity - Privately Held Company, Self, American Psychological Association, Royalties, Self, Guilford Press, Royalties, Self.

P32. BRAIN CONNECTIVITY CHANGES UNDERLYING VIOLENT BEHAVIOR IN A VIDEO GAME AFTER TRANSCRANIAL DIRECT CURRENT STIMULATION: A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, CROSS-OVER TRIAL IN VIOLENT OFFENDERS

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Background: Although no single brain region has been conclusively identified as a causal marker of aggression, disturbances in a network of regions - including the dorsolateral prefrontal cortex (dlPFC) - are consistently observed in individuals prone to violence. This has motivated research into interventions capable of modulating neural circuits underlying aggression. Brain stimulation techniques, such as transcranial direct current stimulation over the dorsolateral prefrontal cortex (dlPFC) can modulate neural circuits implicated in aggression, yet its behavioral and connectivity effects remain unclear.

Methods: In a randomized, double-blind, placebo-controlled, cross-over design, we investigated the neural underpinnings of aggression during a violent video game after anodal stimulation of the dlPFC. 17 male violent offenders (VO) and 17 non-violent controls (CO) completed two sessions (active and sham), followed by MRI acquisition during the video game presentation. We quantified aggressive acts during the game. Functional connectivity analyses examined relationships between in-game aggression, trait measures and stimulation effects.

Results: VO exhibited higher in-game aggression than CO, accompanied by increased functional connectivity among aggression-related regions. Only CO in the first session reduced aggressive behavior after active stimulation along with increased connectivity between dlPFC and right posterior insula. In the VO group, the posterior insula showed positive connectivity towards dlPFC and precuneus related to a higher number of attacks. Positive connectivity between precuneus and posterior insula was as associated with increased reports on traits of aggression, psychopathy, impulsivity and depression in VO. In contrast, more aggressive CO in the video game displayed reduced connectivity within the aggression network. CO with higher trait aggression showed diminished connectivity between centromedial amygdala and posterior insula as well as central opercular cortex.

Conclusions: Our findings reveal distinct functional connectivity mechanisms underlying aggression in violent video games for VO and CO, with the posterior insula emerging as a key hub. tDCS effects on aggression-related networks appear to depend on both individual characteristics and game familiarity, highlighting the complexity of neuromodulation in aggressive behavior.

Keywords: Aggressive behavior, Non-invasive Neuromodulation, Frontal Connectivity

Disclosure: Nothing to disclose.

P33. CIRCUMPLEX MAPPING OF NON-INVASIVE FACIAL EXPRESSIONS PROVIDES OBJECTIVE DEPRESSION SEVERITY AND GROUP CLASSIFICATION

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Background: Digital phenotyping based on facial expression analysis is a promising approach to measure mood and anxiety

severity in the real world that is cheap, low-burden, and scalable. This approach will ultimately help individuals and clinicians monitor and manage symptoms in real-time. However, little work has examined facial features and their dynamics in a naturalistic context in relation to specific clinical symptoms.

Methods: A total of $N = 57$ participants, 46 with major depressive disorder (MDD), and 10 healthy controls (HC), completed audio-visual (AV) recordings during clinical interview (SCID-5). The AV data was then processed via DeepFace, an open-source deep-learning facial recognition algorithm. Probability percentages of facial expression estimates (i.e., Happy, Sad, Fear, Angry, Surprise, and Disgust) were computed per frame, thus creating a time-series for each emotion. Given the multivariate nature of our facial expressions data, we analyzed the data within the framework of the circumplex model of affect, which proposes that affective states emerge from two core neurophysiological systems: one governing valence (ranging from pleasure to displeasure), and the other regulating arousal or alertness. Each participant's time series was projected onto the circumplex model representing the valence-arousal space, using composite scores derived from the weighted contributions of all facial expressions at each time point. To derive quantitative global estimates, each participant was represented with a coordinate within this 2D Valence-Arousal sub-space using a composite score on the mean overall facial expressions across the entire time course. Within this framework, two key sub-space dimension measures can be calculated for each participant; 'Distance'- the Euclidean distance from the center, which indicates how far they are from Neutral (center); and 'Angle'- the direction in radians from the center, which indicate the emotional quadrant the participant is in. Together, these measures provide a quantitative estimate of the individuals' cognitive-emotional state within a given context or timeframe. Two-sample independent t-tests were used to examine group differences in circumplex features: Valence, Arousal, Distance and Angle. Linear regression models were used to examine associations between circumplex features and depression severity (measured by the Quick Inventory of Depressive Symptomatology (QIDS)), while controlling for age, sex, medication use and SCID-5 duration. Machine learning support vector machine (SVM) classification was used for group classification (MDD vs. HC), where Distance and Angle measures were used as the model inputs. We then assessed the generalization ability through fivefold cross-validation.

Results: Participants with MDD showed expected facial emotion profiles of increased Distance ($t(54) = 4.39$, $p = 0.0005$, Cohen's $d = 1.39$), indicating overall more emotive facial expressions, decreased Angle ($t(54) = -2.45$, $p = 0.017$, Cohen's $d = -0.41$), indicating more unpleasant and low arousal expressions, and an overall negative Valence of expressions ($t(54) = -6.20$, $p = 0.00001$, Cohen's $d = -1.45$) compared to HC, controlling for age, sex, medication use and SCID-5 duration. As expected, higher depression severity (QIDS) was significantly associated with increased Distance from center ($R^2 = 0.19$, $p = 0.003$), and more negative Valence ($R^2 = 0.22$, $p = 0.0015$), controlling for age, sex, medication use and SCID-5 duration. Using supervised machine learning SVM classification of group allocation, our models demonstrate that the use of Distance and Angle as predictors achieved 85.7% overall accuracy, with sensitivity of 91.3%, specificity of 60% ROC-AUC of 0.87 and F1-Score of 0.915. While our overall sample is small, yet, the Distance/Angle model outperformed all other sets of predictor variables, such as Valence and Arousal (80.4% accuracy, 87.0% sensitivity, 50.0% specificity, 0.85 ROC-AUC and 0.872 F1-Score), and the 7 basic emotion expression weights (75.0% accuracy, 84.8% sensitivity, 30.0% specificity, 0.71 ROC-AUC and 0.847 F1-Score).

Conclusions: Together, this work demonstrates how objective, computational deep-learning models can be applied to facial expression-based timeseries data in order to derive specific,

individualized assessments of depression severity and potentially identify digital biomarkers for better diagnostics and for prediction of symptom dynamics and trajectories over time, paving the way for more personalized and proactive mental health care.

Keywords: Major Depression Disorder, Automated Facial Affect Recognition, machine learning classification

Disclosure: Nothing to disclose.

P34. SECURE CLOUD-BASED ARCHITECTURE FOR SENSITIVE AUDIO-VISUAL DATA: ENABLING FULL TRANSCRIPTION AND PRIVACY-PRESERVING ANALYSIS

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Background: Raw audiovisual (AV) data resulting from open-ended prompts or clinical interviews are increasingly valuable for clinical and behavioral research. When paired with artificial intelligence (AI) algorithms, these data can yield biomarkers to inform clinical prediction, treatment response, and symptom trajectories. However, direct access to raw AV files raises significant concerns about privacy, data security, and regulatory compliance. There is a critical need for infrastructures that allow sensitive AV data to be processed safely and effectively, while ensuring that researchers can benefit from derived information without unnecessary exposure to protected health information (PHI).

Methods: We describe a cloud-based architecture designed to process sensitive AV files in a secure and compliant manner. The architecture incorporates multiple layers of protection: (1) encrypted storage and transfer of raw data; (2) strict access controls ensuring that only authorized personnel can interact with identifiable files; and (3) an isolated environment where AI-based transcription and analytic workflows can be applied safely without exposing raw recordings. Two complementary pipelines are developed: first, automated transcription using state-of-the-art speech-to-text models with integrated personal identifier (PII) redaction, providing full-length transcripts that can be benchmarked against human-edited outputs; and second, a privacy-preserving analytic framework enabling researchers to access derived features (linguistic, prosodic, facial metrics) without direct exposure to raw AV data.

Results: This architecture is expected to meet established security and compliance standards (e.g., HIPAA, GDPR) through layered encryption, role-based access, and isolated compute environments. Flexibility in design allows for scalable storage and computational resources, ensuring the system can adapt to growing data volumes and evolving analytic needs. In addition, the architecture is designed to support efficient resource allocation, enabling compute power and storage to be scaled up or down as needed to optimize performance and cost. This infrastructure is anticipated to support both automated transcription and secure feature extraction pipelines while maintaining strict privacy protections.

Conclusions: We outline a secure, scalable infrastructure for processing sensitive AV data and enabling AI-based analyses in clinical and behavioral research. By balancing the need for data accessibility with stringent security and regulatory requirements, such architectures can responsibly expand the use of AV-based digital biomarkers while safeguarding participant privacy. This model can be broadly applied across research domains to maximize scientific value while maintaining ethical and legal standards.

Keywords: AI, Automated natural speech analysis, neuroinformatics, regulatory compliance, architecture

Disclosure: Nothing to disclose.

P35. DATA-DRIVEN SUBTYPES OF MAJOR DEPRESSIVE DISORDER (MDD) SHOW VARIABLE TREATMENT RESPONSE TO SERTRALINE

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Background: Major depressive disorder (MDD) is a major public health burden worldwide. The disorder features high heterogeneity in clinical symptomatology, neurobiological underpinnings, and subsequent treatment response. Only half of MDD patients respond to first line SSRI treatment, and those non-responding patients often try several additional treatments to improve response, as evaluated through large and complex studies like STAR-D [Rush 2006]. Patient-level clinical and neurobiological predictors of antidepressant treatment response could enable higher probabilities of remission with fewer treatments. Personalized treatment allocations require extensive treatment response data across multiple mechanisms of drug action in diverse patient populations. However, the first step on this path involves identifying reproducible MDD patient subtypes, established prior to treatment initiation, that show variable treatment response to a single drug like sertraline.

Methods: We performed unsupervised uni- and multi-modal clustering using baseline/pre-treatment data in the Establishing Moderators and Biosignatures of Antidepressant Response in Clinical Care (EMBARC) sertraline clinical trial dataset [Trivedi 2016] using selections of clinical and behavioral data (N = 294, 113 features), resting and evoked electroencephalogram (EEG) data and various human neuroimaging (MRI) data. Human neuroimaging data (structural: N = 278, resting state functional: N = 263, and task-based functional: N = 228 magnetic resonance imaging; MRI, rs-fMRI, and tb-fMRI, respectively) from baseline (pre-treatment) and week 1 (post-treatment) were processed using Human Connectome Project workflows in QuNex [Demšar 2023]. We produced 232 structural/anatomical MRI features, 946 rs-fMRI features and 1008 features across two tasks of tb-fMRI. Human EEG (N = 203 subjects, 2444 features across resting and evoked states) were processed using MNE [Gramfort 2013]. Uni- and multimodal clustering of patients was performed using baseline data with [multimodal] variational autoencoders ([M]VAEs) followed by spectral clustering of resulting embeddings [Banerjee 2023]. We performed seven unimodal clustering and two multimodal clustering (clinical features with all imaging or just rs-fMRI features) analyses. Cluster stability was assessed within baseline/training data by cross-validation using adjusted mutual information (AMI) [Vinh 2010]. Placebo-adjusted treatment effects of sertraline on HAMD-17 were calculated using mixed models for repeated measures (MMRMs) [Mallinckrodt 2001], and variation in placebo-adjusted treatment response by cluster was assessed using ANOVA.

Results: We performed seven unsupervised unimodal clustering (clinical, EEG, anatomical, structural/thickness, inter- and intra-network, functional connectivity and task contrasts) and two multimodal clustering (clinical + all imaging, clinical + rs-fMRI) analyses in the EMBARC baseline data. Clinical-only clustering identified four MDD clusters (AMI = 0.87) that differed on overall symptom severity and variation in symptom subdomains related to appetite and sleep. These clusters showed significant

differences in treatment response (ANOVA $p=0.018$), with two clusters showed enhanced treatment effects (cVAE_C11: LSMD -4.3 , $p=0.008$, $d=-1.15$; cVAE_C14: LSMD -3.5 , $p=0.019$, $d=-0.94$) compared to the overall study population (LSMD 1.2 , $p=0.14$, $d=-0.33$), and two clusters, both with lower disease severity, showed enhanced placebo effects (cVAE_C12: LSMD $+1.2$, $p=0.44$, $d=0.32$; cVAE_C13: LSMD $+1.6$, $p=0.40$, $d=0.44$). Unimodal clustering of neuroimaging and EEG data all produced between 3 and 5 clusters, and each showed high clustering stability. However, the resulting clusters showed little variation in (held-out) clinical/behavioral data, and little overlap when computing the pairwise AML across modalities. Nevertheless, clusters produced by rs-fMRI functional connectivity and combined resting state and evoked EEG showed significant variation in treatment response (ANOVA $p=0.004$ and 0.014 respectively), while clusters created from structural and task-based neuroimaging features did not (ANOVA p -values: 0.41 – 0.74). Variation in treatment response was largely driven by placebo-responsive EEG cluster 3 (LSMD $+3.9$, $p=0.053$, $d=+1.0$), which featured increased peak height and peak width coherence in loudness dependence of the auditory evoked potential (LDAEP), and sertraline-responsive rs-fMRI functional connectivity cluster 4 (LSMD -8.6 , $p=1e-4$, $d=-2.3$) which featured connectivity differences most prominent in somatomotor, temporal cortices, and anterior thalamus. While multimodal clustering with clinical and neuroimaging data successfully produced stable clusters, neither set showed significant variation with treatment response (ANOVA $p=0.54$ and $p=0.16$, respectively).

Conclusions: MDD patient clustering based on clinical or neurobiological characteristics, generated prior to treatment, identified subpopulations that subsequently showed differential responses to SSRIs. Future work could evaluate whether patients in clusters that performed poorly on SSRIs might respond better to other mechanisms of action. These results together highlight unsupervised strategies for advancing precision medicine in MDD.

Keywords: Machine learning clustering, Major Depressive Disorder (MDD), Human Neuroimaging, EEG biomarkers

Disclosure: Neumora Therapeutics, Employee, Self.

P36. GENERATING AUTOMATED RATINGS FOR THOUGHT DISORDER THROUGH FINETUNING A LARGE LANGUAGE MODEL (LLM)

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Background: Thought disorder is a cross-diagnostic symptom dimension with substantial implications for functional outcomes. Our objective was to adapt a Large Language Model (LLM) to automatically predict the severity of several aspects of thought disorder based on transcribed speech.

Methods: Speech was recorded and transcribed from 592 participants across multiple diagnoses, including healthy volunteers and individuals with anxiety, mood disorders, and psychosis. Recordings were made while participants responded to language tasks, including fluency tasks (animal and f-letter), picture descriptions, and open-ended narrative prompts. Documents were split by stimulus, generating 7585 samples. Participants were also evaluated clinically by a trained assessor, and ratings for thought disorder were completed using the Scale for the Assessment of Thought, Language, and Communication (TLC). The following TLC items were selected for prediction based on their prevalence, ability to be rated reliably, and span of positive and negative thought disorder: poverty of speech, poverty of

content of speech, distractibility, derailment, circumstantiality, perseveration, loss of goal, incoherence, neologisms, tangentiality, pressured speech, and illogicality.

The samples were split into 90% training and 10% test sets. The training set was used to fine-tune GPT-4o via low-rank adaptation ("Fine-tuned Approach"). Model performance in the test set is represented using accuracy and mean absolute error (MAE). Performance was benchmarked against a zero-shot approach utilizing the native GPT-4o model with no fine-tuning ("Prompt Approach") as well as an approach where examples of the thought disorder phenomena were provided ("Enriched Prompt Approach").

Results: The fine-tuned approach achieved high agreement with human annotators and low distance from correct judgments, on average achieving MEA of 0.55 points from the clinical rating, compared to MEA of 1.6 in the Prompt Approach, and MEA of 1.4 in the Enriched Prompt Approach. The fine-tuned model also improved task-independent accuracy to 62% from 10 to 12% in the other approaches. The improved performance was present across all TLC items, and for all of the task stimuli. For the TLC items, the lowest MEA was achieved in identifying neologisms, followed by incoherence and poverty of speech. Performance was relatively even across the language task stimuli.

Conclusions: We demonstrate that fine-tuning an LLM on transcribed speech can generate relatively accurate automated ratings for thought disorder, and that this approach is superior to prompt-based approaches. This approach may provide an avenue for scalable quantification of illness severity in clinical applications.

Keywords: Large Language Models, formal thought disorder, psychosis

Disclosure: Cogntiv, Consultant, Self, North Shore Therapeutics, Founder, Self, North Shore Therapeutics, Board Member, Self, Psyrin, Advisory Board, Self.

P37. THE VOICE-ENABLED VIRTUAL PATIENT: A PILOT STUDY ON SYSTEM PERFORMANCE AND CLINICAL VALIDITY

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Background: Training mental health clinicians to conduct standardized diagnostic interviews is challenging due to a lack of scalable, realistic practice opportunities. Traditional methods often fail to prepare trainees for the variability and complexity of real-world patient interactions, potentially impacting data quality in clinical trials. Here, we introduce a novel approach to address this training gap using Large Language Model (LLM)-based interview simulations to produce a voice-enabled virtual patient simulation system. In this proof-of-concept study, we introduce the methods for developing this tool and collect pilot data with expert raters to assess whether this voice-enabled virtual patient achieves the following three goals: (1) accurately adhere to pre-defined clinical profiles and symptom severity, (2) maintain a coherent and consistent narrative, and (3) produce dialogue that is perceived as realistic.

Methods: We implemented a system that uses an LLM to power interactive, voice-enabled virtual patients with specified symptom profiles, demographic details and backstories, and distinct communication styles. In our pilot study, initial clinical validity was evaluated by experienced clinical raters who conducted Montgomery-Asberg Depression Rating Scale (MADRS) structured interviews with four unique virtual patient personas. We configured four fixed clinical profiles ("Susan", "Brian", "Elena", and "David") that each portrayed a different level of symptom severity,

unique backstory, and either a forthcoming or more guarded communication style. Raters conducted MADRS interviews on each of the four personas. Adherence to clinical profiles was assessed by comparing raters' scores with programmed scores and inter-rater reliability was calculated between raters for each of the four personas. Raters were then asked Likert-style questions pertaining to clinical plausibility, narrative coherence, and dialogue realism of each interaction.

Results: Initial results (n interviews = 12) show that the virtual patients demonstrated strong adherence to their configured clinical profiles, with human raters scoring the patients with high accuracy against their ground-truth score configurations. The mean item difference between rater scores and configured scores was 0.38 (SD = 0.76). The item that demonstrated the highest adherence was Suicidal Thoughts (mean item score difference = -0.08, SD = 0.51) and the lowest adherence was Reduced Appetite (mean item score difference = 0.75, SD = 0.97). The interclass correlation coefficient for inter-rater reliability across items was 0.90 (95% CI = 0.34–1.0). Expert raters also found the virtual patients to be qualitatively realistic and cohesive with average ratings of "Agree" to "Strongly Agree" in response to statements asking how much raters agreed that the virtual patients performed well in these areas.

Conclusions: LLM-powered virtual patient simulations represent a viable and scalable new tool for training clinicians in standardized diagnostic interviewing and scale administration. This proof-of-concept pilot study demonstrates the system's ability to produce high-fidelity, clinically relevant practice scenarios. Future work will focus on rigorously assessing the system's impact on trainee performance over time.

Keywords: Generative Artificial Intelligence, MADRS, Education and Training

Disclosure: Brooklyn Health, Founder, Self.

P38. USING LARGE LANGUAGE MODELS FOR ENDPOINT OVERSIGHT IN PSYCHIATRIC DRUG DEVELOPMENT

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Background: Large Language Models (LLMs) have the potential to provide oversight of ClinRO endpoints due to their ability to process text and, through extensive training, learn to accurately infer and assign a score to language-based sentiment. We created Hammy- a system of LLMs which transcribe ClinRO interviews, parse out individual ClinRO items, and provide associated scoring. Here we outline the methodology used to develop and train Hammy and discuss using it to perform a post-hoc data quality check on Hamilton Anxiety Rating Scales (HAM-A) from a recent Phase 2b clinical trial. Finally, we discuss the implications of deploying this technology for data monitoring in ongoing clinical trials.

Methods: Hammy consists of multiple concurrent models that ingest audio recordings of ClinRO interviews and produce associated scores for each item of that interview. The first step utilizes Whisper, an open-source transcription model which we prompt engineered to more accurately transcribe ClinRO interviews. After audio recordings are transcribed by the model, a second model parses the transcripts into item-level segments. A third model analyzes this version of the transcript and produces a score for each item. These models are based on Llama 3.1 and were trained on data from a recent Phase 2 clinical trial run by Mind Medicine, Inc (MindMed), where over 1500 HAM-A interviews were audio recorded. Once trained, a cross-

validation technique was used to create different versions of Hammy in order to appropriately test performance on the Phase 2 dataset as well as approximate scoring confidence in other datasets. As a measure of performance, Hammy scored three gold standard training interviews used to certify human raters. Finally, we re-analyzed the results of the Phase 2 study using Hammy's scoring in place of the original central rater scoring.

Results: On average, Hammy's scores differed 1.57 (+/- 1.39) points from the central raters' scores, with Pearson's $r = 0.98$ indicating a very strong relationship between the two sets of scores. On training interviews, Hammy matched the "answer key" scoring for all 14 items (100%) on the first 2 recordings and for 12 of the 14 items (85.7%) for the third recording. Hammy's scoring reaffirmed the topline results of the Phase 2b trial. Notably, Hammy's scoring would have resulted in 21 participants who were originally excluded from the study based on HAM-A scoring below to be included and 7 participants who were originally included to be excluded. Exclusion of these 7 participants would not have significantly changed results.

Conclusions: Our work demonstrates how LLMs can be trained to score ClinRO assessments. Hammy's scores on the HAM-A are comparable to human raters, and well within normal levels of inter-rater variability (CITE). Hammy consistently matched the "answer key" on a series of central rater training videos, scoring well above the threshold needed to receive certification. This analysis indicates that LLMs have the potential to be deployed as a consistent and relatively inexpensive method of providing oversight of clinical outcomes ratings in trials. As LLMs deploy a uniform scoring methodology and can rapidly score every interview conducted, they can provide a level of data quality oversight that has previously been prohibitively expensive or logistically impossible to obtain via human raters. Future directions of this work include applying this methodology to other scales, expanding the pool of training data with different raters, and using Hammy for near real-time rater oversight in active clinical trials.

Keywords: precision psychiatry, artificial intelligence, Endpoint Reliability

Disclosure: Mind Med, Employee, Self.

P39. GEOMETRIC FEATURE-BASED MACHINE LEARNING FOR REPRODUCIBILITY OF 18F-FLORZOLOTAU PET STAGING LABELS IN REPETITIVE TRAUMATIC BRAIN INJURY PATIENTS

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Background: Repetitive traumatic brain injury (TBI) is associated with chronic traumatic encephalopathy (CTE), a neurodegenerative disease presenting behavioral and mood changes, cognitive impairment and dementia. The neuropathological stages of CTE have been solely determined in postmortem examinations, where hyperphosphorylated tau protein initially deposits in the sulcal depths and progressively spreads across widespread cortical regions. Although early diagnosis and intervention for CTE are urgently needed, no non-invasive method for staging CTE has yet been established. In this study, we propose novel tau staging labels on 18F-Florzolotau positron emission tomography (PET) images, which may correspond to the neuropathological staging framework proposed by McKee et al. (2013). The three imaging-based categories are: No Tau, Focal Sulcal Tau

Pathology, and Extensive Cortical Tau Pathology. The aim of this study is to apply machine learning to evaluate the reproducibility of these staging subtypes and to characterize their imaging signatures using geometric features derived by standard radiomics approaches.

Methods: A total number of 105 individuals under the age of 85 without clinically apparent dementia or late-onset psychiatric disorders were included, of which 45 had a history of repetitive sports-related TBI (mean age 45.9 ± 12.0 years, 42 male [93%]) and 48 healthy controls (HC) (mean age 45.0 ± 10.0 years, 28 male [58%]). All participants underwent 18F-florbetapir PET imaging up to three times to examine longitudinal patterns of tau deposition. SUVR maps were generated using a data-driven gray matter reference region derived from histogram fitting proposed in Tagai et al. (2022). All PET scans were spatially normalized to a common template using SPM. Voxel-wise t-maps were generated for each TBI subject by comparing SUVR values against the mean SUVR of the HC group. Tau clusters were defined as contiguous voxels (>5 voxels) with $t > 2.0$. Cluster geometry was quantified by applying the Marching Cubes algorithm to estimate volume, surface area, and surface-to-volume (S/V) ratio. These geometric statistics were summarized as feature vectors for each subject. We then trained binary and multinomial logistic regression models to classify PET images into the three proposed staging labels (no tau, focal sulcal tau pathology, and extensive cortical tau pathology). Model performance was evaluated using nested 5-fold cross-validation, with the area under the curve (AUC) and classification accuracy reported. Feature importance analyses were further conducted to identify geometric characteristics contributing to stage discrimination. Visual inspection of misclassified cases was performed to consider any methodological fallacy. We also considered the possibility that the cerebellar region may be susceptible to tracer off-target binding. Therefore, cerebellar masks were created based on the Desikan–Killiany atlas (Freesurfer) from individual T1-weighted MRI, and the entire analysis was conducted with and without excluding cerebellar regions.

Results: The average AUCs of the binary and multinomial models for three-class classification were 0.881 ± 0.107 and 0.903 ± 0.061 , respectively, with mean classification accuracies of $81.4 \pm 12.2\%$ and $68.2 \pm 7.5\%$. Extensive Cortical was primarily characterized by the maximum and median values of cluster surface area, whereas Focal Sulcal was distinguished by the number of tau clusters and the maximum surface-to-volume (S/V) ratio. No Tau was identified based on a composite of multiple geometric parameters. In some misclassified cases, visual inspection indicated that the staging labels predicted by machine learning from $t > 2.0$ maps appeared more consistent with the observed tau patterns than the original read-off labels. When cerebellar regions were excluded, the average AUCs of the binary and multinomial models lowered to 0.872 ± 0.127 and 0.883 ± 0.072 , with mean accuracies of $80.9 \pm 12.7\%$ and $72.6 \pm 4.3\%$, respectively, suggesting that cerebellar exclusion did not materially alter the classification performance.

Conclusions: We developed classifiers that reproduced tau PET-based staging with relatively high accuracy, supporting the reproducibility of imaging-derived labels for non-invasive CTE staging. Our findings further suggest that automated classification may provide an unbiased tool for non-invasive assessment of tau accumulation patterns independent of rater variability. Future studies should probe the concordance between these imaging-based staging labels and the conventional neuropathological staging and aid the development of an early, non-invasive diagnosis protocol for CTE.

Keywords: Traumatic Brain Injury, F-18 PET Imaging, machine learning classification, Artificial Intelligence, Analysis, Neuroscience, Clinical Neuroscience, Pharmacology, Dementia-related psychosis

Disclosure: Nothing to disclose.

P40. CREATING A FRAMEWORK FOR LARGE LANGUAGE MODELS FOR CAREGIVER SUPPORT IN DEMENTIA

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Background: Caregiving in dementia is associated with a range of negative outcomes including worse mental health (depression, anxiety), caregiver burnout, financial strain and higher mortality. Geriatric care managers (GCMs) are a major resource for solutions, including (i) providing information (ii) enhancing caregiver competency, and (iii) providing emotional support. However, GCM services are not typically covered by insurance and are not easy to access because of a shortage of GCMs. Thus, there is a need for solutions to fill this gap. The recent emergence of large language models (LLMs) such as ChatGPT that can provide information, support, and potentially guidance offers tantalizing possibilities. The increasing ubiquity of LLMs means that it is reasonable to anticipate that caregivers will increasingly turn to these tools for a range of solutions.

However, there are major unanswered questions, including the absence of any evidence base or meaningful best practices governing LLM use. We aim to identify specific domains of care management that may be best served by AI versus GCMs to help begin to answer these concerns.

Methods: We recruited 25 caregivers of persons with dementia. Caregivers ranged in age from 22 to 83 years, with 64% identifying as female, and included both family and paid caregivers. We conducted five focus groups with caregivers, generating a list of more than 100 common questions, categorized into factual, emotional, and decision-making domains. We chose 75 of these questions (25 of each domain subtype) and posed each question independently to a dementia-specific large language model (LLM) developed by Rippl and to our study clinician (Marie Clouqueur, LICSW) who is a geriatric care manager (GCM). The queries were then re-posed to our GCM while giving her the ability to review the responses generated by the LLM. This in turn produced three answers to each of the 75 caregiver questions, one written by the LLM, one by the GCM, and one by the GCM alongside the LLM. Subsequently, we asked our caregiver cohort to blindly rank the three responses for each question, with each caregiver assigned 15 questions to assess. Our statistical analyses will determine which responses caregivers preferred overall and by question domain subtype.

Results: With the study completed in July 2025, primary quantitative findings are not yet available at time of this submission but will be ready for presentation by the Annual Meeting in January 2026. Qualitatively, we found a stark difference in response times between the Geriatric Care Manager (GCM) and the large language model (LLM) when composing answers to the 75 questions. The LLM's quick speed underscores its potential value—both in providing caregivers with immediate access to information and in saving time for GCMs. Additionally, our study used one-shot responses from the LLM, so we asked the GCM to respond in the same manner for consistency. Our GCM qualitatively reported that she would have much preferred the ability to have a back and forth dialog with the caregiver to answer the queries. This clearly demonstrates that when it comes to caregiver support, the context of an interaction between a caregiver and human or AI resource is important. Not all caregiver questions are well suited to a single-response format, and important conversational elements—such as a GCM's ability to express empathy and assess caregiver distress—may be lost in a one-shot LLM exchange.

Conclusions: This study is a first step towards better understanding the ability for LLMs to play a role in caregiver support similar to that of geriatric care managers. Through our analyses, we will determine which question domain subtypes (factual, emotional, decision-making) may be best served by a large language model and which may be best answered by a human. We will also report qualitative and quantitative data from additional survey results that may guide meaningful interactions between caregivers and large language models.

Keywords: Dementia Caregiving, Large Language Models, Caregiver Support Chatbot

Disclosure: Nothing to disclose.

P41. HETEROGENEITY IN THE CAUSAL FACTORS FOR PAIN INTERFERENCE IN ADULTS WITH SPINAL CORD INJURY-RELATED NEUROPATHIC PAIN

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Background: About 69% of the 308,620 Americans with spinal cord injury (SCI) suffer from debilitating chronic neuropathic pain, significantly impacting functional ability and quality of life. Treatment options are mainly limited to medications, which often provide insufficient benefits and carry risks for addiction and side-effects. Many variables can influence chronic pain, with stress, anxiety and depression affecting about 60% of adults with SCI-related neuropathic pain. Those more concerned about their pain typically experience a stronger connection between pain intensity and its interference in daily activities. Given the heterogeneity in pain interference, understanding individual differences in contributing holistic variables is crucial. Ecological momentary assessment (EMA) surveys can measure pain interference as a dynamic process as participants provide real-time data about daily experiences and behaviors in their natural environment. To identify the individual causal relationship between psychosocial variables and pain interference, we applied causal discovery analysis to EMA surveys from persons with chronic neuropathic pain from SCI.

Methods: This is a remote observational study. Participants were adults with SCI sustained ≥ 1 year ago, medically stable, and neuropathic pain severity of ≥ 4 on the Numerical Pain Rating Scale over the past week. Exclusion criteria were cognitive impairment and/or communicative disability (e.g., due to brain injury) preventing them from following directions and major medical complications. EMA surveys were collected 4x/day for 21 days and encompassed questions about sleep, pain intensity and interference with function, positive and negative affect, stress, body/mind awareness, agency (i.e., control over one's pain and life), and two brief cognitive tests. Causal discovery analysis, a machine learning method that searches for causal relationships among variables in an observational dataset, was applied to the data to create a causal graph for each participant. The frequency of causal factors for pain interference was quantified and visualized.

Results: We recruited 17 adults (8 men) with SCI-related neuropathic pain. They were on average 46 ± 17 years old (range 22–77 years). Nine participants had a cervical lesion (tetraplegia); 8 had a thoracic lesion (paraplegia). They were 11 ± 9 years (range 1–33 years) post-SCI. Their highest neuropathic pain was on average 7 ± 1 on the Numeric Pain Rating Scale (ranging from 0 = no pain to 10 = worst pain ever). For 15 participants: pain intensity, concern over pain, and fear of moving because of pain were causal for increased pain interference. For 10 participants: spatial awareness, being active, determined, attentive, inspired,

alert, having agency (control over life or pain), and/or having a relaxed mind and body were causal for increased pain interference. For 6 participants, agency (control of pain, control of life) was causal for reduced pain interference.

Conclusions: Several of the variables found in the causal discovery pathways are modifiable, for example, concern over pain, fear of moving because of pain and having agency. Mind and body approaches such as Qigong (i.e., mindful movement) can increase agency, awareness of body and mind, and reduce fear and concern for pain. Further studies would need to be performed to investigate whether these individual causal pathways for pain interference are modifiable with mind and body approaches.

Keywords: ecological momentary assessment, causal discovery analyses, neuropathic pain

Disclosure: Nothing to disclose.

P42. A ROBUST LLM FRAMEWORK FOR ASSESSING EXTERNAL CONTRIBUTORS TO PSYCHOSIS RISK IN CLINICAL HIGH-RISK INDIVIDUALS FOR SCHIZOPHRENIA

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Background: To better understand the factors influencing symptom progression in individuals at clinical high risk for schizophrenia (CHR), we developed a structured framework for assessing external contributors to psychosis risk grounded in clinical literature and patient narratives in open-ended interviews. The unstructured interview setting provides spontaneous insights into individuals' inner experiences and their connection to external factors, such as the impact of childhood trauma. Our framework organizes these contributors into three categories: chronic vulnerabilities (e.g., childhood trauma, long-term social isolation, poverty, discrimination), acute stressors (e.g., romantic conflict, housing loss, substance use escalation), and warning signs observable in patient expression (e.g., emerging paranoia, disorganized thought, affective blunting, withdrawal). This categorization creates a clinically meaningful codebook for systematically analyzing interviews and identifying factors that may precede or drive psychiatric episodes.

Methods: We tested this approach using data from the Accelerating Medicines Partnership® in Schizophrenia (AMP-SCZ) study, including 711 participants (HC: 134, CHR: 578) across 24 sites. The mean age was 22.1 ± 3.6 years for HC and 21.0 ± 4.1 years for CHR participants. Positive symptoms were assessed with the PSYCHS-SIPS as a proxy of CHR severity. All participants completed open-ended interviews in English, which were recorded and transcribed using TranscribeMe.

Transcripts were analyzed with the SOLOMON framework, a reasoning system that enhances large language model (LLM) reliability by systematically exploring solution spaces, integrating diverse perspectives, and synthesizing unified responses. In the exploration stage, multiple instruction-tuned LLMs—Granite-3-3-8B, Granite-3-2-8B, Llama-3-3-70B, and Llama-3-405B—independently scored and extracted evidence for the evaluated external factors. Their outputs were reviewed in the aggregation stage by a judge model, Llama-4-Maverick-17B, which consolidated results into a consensus to ensure both breadth of reasoning and robustness. These final scores were used as features for analysis purposes.

Group comparisons (HC vs CHR) were performed on these scores using the Mann–Whitney U test, with p-values adjusted for multiple comparisons using FDR correction. Then, these scores were used to predict PSYCHS-SIPS positive total scores using ridge regression with 10-fold cross-validation. Outcome measures included concurrent symptoms (± 15 -day window from the interview date) and future assessments (60–90 days from the interview date). In addition, Shapley Additive exPlanations (SHAP) analysis was applied to assess the contributions of these external factors to the predictions. This approach enhances model interpretability by quantifying the impact of each factor, providing both methodological and empirical insights into symptom progression.

Results: The largest differences between HC and CHR participants were observed for neurodevelopmental or cognitive issues, social withdrawal, and chronic isolation, or poor social support (all $p < 0.00001$), highlighting chronic vulnerabilities and enduring psychosocial risk factors. Using a cross-validated regression approach, Pearson correlations between predicted and observed PSYCHS-SIPS scores were moderate for both current assessments ($n = 439$; $r = 0.41$, $p < 0.00001$) and future assessments ($n = 187$; $r = 0.39$, $p < 0.00001$). SHAP analysis indicated that top contributors to current scores were primarily chronic vulnerabilities (e.g., patients talk about the difficulties with the performative nature of conversations and describe struggles with self-expression and social interactions) and warning signs. For future scores, the most influential features shifted toward acute stressors, such as job loss/financial stress and eviction or sudden moves (e.g., patient described being fired due to illness, struggling to secure employment despite numerous applications, and facing financial stress from costly schooling and unstable housing), suggesting that acute psychosocial triggers become increasingly relevant in forecasting symptom progression over time.

Conclusions: The proposed feature categorization, combined with LLM-based analysis, offers a robust framework for identifying and quantifying external contributors to symptom progression. Our results align with existing clinical findings, confirming that chronic vulnerabilities and warning signs play a dominant role in current symptom severity among individuals at clinical high risk for schizophrenia, serving as stable markers for early detection. Moreover, we observed that acute stressors significantly influence short-term symptom trajectories, underscoring the importance of monitoring both persistent and dynamic contributors. In this way, our method provides valuable insights into the relationships and evolving patterns of symptom progression.

Keywords: Large Language Models, Clinical high-risk for psychosis, Disease risk prediction

Disclosure: Nothing to disclose.

P43. SENTIMENT ANALYSIS OF A 324-SESSION PSYCHOANALYTIC PSYCHOTHERAPY COURSE REVEALS GRADUAL INCREASE IN POSITIVE EMOTION IN PATIENT STATEMENTS

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Background: Psychotherapy remains a cornerstone of modern mental health care; for common conditions such as depression and anxiety, evidence suggests that psychotherapy often demonstrates higher effect sizes than antidepressant monotherapy. Despite its clinical importance, monitoring therapeutic progress in real time remains labor-intensive, reliant on detailed process notes, expert coding, or retrospective chart review. As a result, identifying specific mechanisms of change across the course of

psychotherapy has been challenging. Recent advances in natural language processing (NLP) and deep learning offer new tools for quantifying emotional, cognitive, and interpersonal processes embedded in session dialogue. These methods may provide scalable approaches for measuring progress within a course of treatment and for identifying which therapeutic elements most directly contribute to positive outcomes.

Methods: We conducted a computational analysis of a high-frequency psychoanalytic psychotherapy conducted four times per week over the course of 324 sessions. Audio recordings were transcribed and manually de-identified by collaborating investigators prior to analysis. A fine-tuned RoBERTa sentiment analysis model was applied to each talk turn, separately for patient and therapist. Sentiment scores were averaged at the session level and analyzed longitudinally across the treatment course. Windowed averaging was also applied to reduce noise and reveal temporal trends.

Results: Therapist sentiment remained predominantly neutral across the treatment, consistent with the psychoanalytic techniques of the era in which the case occurred. In contrast, the patient's language demonstrated a gradual increase in positive sentiment and a corresponding decrease in negative sentiment across the treatment course. This trend was most clearly visible using windowed averaging, which highlighted a sustained shift toward more positively valenced expression over time. These findings are consistent with the idea that emotional tone in patient discourse may serve as a marker of therapeutic progress.

Conclusions: This case study demonstrates the feasibility of applying modern NLP methods to archival psychotherapy transcripts. Sentiment analysis successfully captured longitudinal improvement in patient affective expression, aligning with expected therapeutic change. While these results highlight the promise of sentiment analysis as a computational marker of clinical progress, they also underscore the need for more refined computational phenotypes—such as measures of self-efficacy, defensive functioning, and interpersonal dynamics—before reliable and generalizable measurements can be made. Future research incorporating larger samples, diverse therapeutic modalities, and integration with clinical outcome data will be essential for validating these methods and for advancing computational psychiatry as a tool to complement traditional psychotherapy research.

Keywords: Psychotherapy, natural language processing (NLP), longitudinal analysis

Disclosure: Nothing to disclose.

P44. AN EEG BIOMARKER PREDICTING PLACEBO RESPONSE IN MAJOR DEPRESSIVE DISORDER: DEVELOPMENT, PROSPECTIVE VALIDATION, AND IMPLICATIONS FOR ENHANCING DETECTION OF TREATMENT EFFECTS

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Background: Placebo response poses a significant challenge in clinical trials for major depressive disorder (MDD), obscuring true drug effects. Biomarkers that predict placebo responses could improve trial efficiency. Here, we developed and prospectively validated an electroencephalography (EEG)-based biomarker of placebo response in MDD and evaluated its utility in augmenting the precision of detecting treatment effects.

Methods: Drawing on the expectation that placebo effects are common to all treatments, we initially trained our biomarker using

resting-state EEG data from multiple open-label trials (N = 589) involving FDA-approved antidepressants (N = 260), repetitive transcranial magnetic stimulation (rTMS; N = 252), and agomelatine (N = 77). The biomarker, optimized through cross-validation, was then validated externally in a Phase 2a open-label trial of the investigational antidepressant ALTO-100 (N = 135) and the EMBARC randomized controlled trial (RCT) comparing sertraline (N = 83) and placebo (N = 99). Subsequently, prospective validation was conducted using placebo-arm data (N = 111) from the ALTO-100 Phase 2b RCT. We further assessed whether incorporating biomarker-based placebo predictions as sample weights could enhance drug-placebo effect sizes using mixed models for repeated measures (MMRM).

Results: The EEG biomarker demonstrated significant predictive validity across diverse datasets. In initial training, it showed a modest but significant correlation with treatment response ($r = 0.09$, $p = 0.012$), which was replicated in external validations: ALTO-100 open-label ($r = 0.15$, $p = 0.041$) and the EMBARC RCT (sertraline: $r = 0.19$, $p = 0.040$; placebo: $r = 0.31$, $p < 0.001$). Prospective analysis of the ALTO-100 RCT placebo arm yielded significant partial correlations with MADRS response at weeks 2 ($r = 0.29$, $p = 0.001$), 4 ($r = 0.24$, $p = 0.006$), and 6 ($r = 0.19$, $p = 0.029$). Incorporating biomarker predictions into MMRM analyses notably increased drug-placebo effect sizes from 0.13 to 0.29 in ALTO-100 RCT and from 0.19 to 0.26 in EMBARC.

Conclusions: Our EEG-based biomarker predicts placebo response across multiple independent trials and enhances precision in detecting therapeutic effects. Integrating EEG-predicted placebo response into analysis may improve the accuracy of antidepressant efficacy assessments in future clinical trials.

Keywords: Placebo Response, Predictive Biomarker, MDD, Electroencephalography (EEG), machine learning

Disclosure: Alto Neuroscience, Employee, Self.

P45. ENHANCING DIGITAL COGNITIVE PROFILING IN SCHIZOPHRENIA AND DEPRESSION WITH SPEECH-DERIVED FEATURES: TOWARD SCALABLE BIOMARKERS IN PSYCHIATRY

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Background: Cognitive impairment is a core and enduring feature of numerous psychiatric disorders, notably schizophrenia (SZ) and major depressive disorder (MDD). Deficits in verbal memory, processing speed, and executive functioning may persist independently of acute symptoms and strongly predict functional outcomes. Despite their relevance, cognitive assessments are rarely implemented in routine psychiatric care due to constraints in time, cost, and ecological validity.

In neurodegenerative disorders like Alzheimer's disease, adding automated speech analysis to cognitive tasks has improved the sensitivity of traditional cognitive assessments in their ability to detect the disease and predict cognitive decline. Early evidence suggests this approach may also be applicable in psychiatric disorders. Building on this, this study explores the potential added value of speech analysis in enhancing cognitive assessment in individuals with SZ, MDD, and healthy controls (HC), with the goal of furthering the development of digital biomarkers for psychiatry.

Methods: Participants with SZ, MDD, and HC were recruited from the Department of Psychiatry, Karl Jaspers Clinic, University of Oldenburg, Germany. Diagnoses were made according to DSM-5; symptom severity was assessed with the Positive and Negative Syndrome Scale (PANSS in SZ) and Montgomery-Åsberg

Depression Rating Scale (MADRS in MDD). All participants completed the smartphone-administered Rey Auditory Verbal Learning Test (RAVLT), consisting of 4 trials with 15 words each (no delayed recall) and the Semantic Verbal Fluency test (SVF, animal naming within 60 s). Verbal responses were automatically transcribed and analyzed using natural language processing, generating 70 features capturing semantic, temporal, and task-specific speech patterns, such as learning slopes. These informed subdomain scores for memory, executive function, and processing speed, as well as a composite cognitive score. Subdomain and composite scores were adjusted for age, sex and years of education using a normative sample. A priori power analysis using G*Power indicated that a sample of 66 participants (22 per group) would be sufficient to detect medium effect sizes ($\eta^2 \approx 0.06$) for group comparisons using the Kruskal-Wallis test, with $\alpha = 0.05$ and power = 0.80. Group comparisons were conducted via Kruskal-Wallis tests. For classification, machine learning models, including Decision Trees (DT), Random Forests (RF), Linear Models (LM), and others, were trained to distinguish diagnostic groups using (1) raw RAVLT and SVF scores and (2) raw test scores with additional speech-derived features. Classifier performance was evaluated using 10-fold cross-validation and AUC as the primary metric; the best-performing models are reported here. Permutation feature importance was used to identify the most informative features.

Results: The final sample included 67 participants (HC: $n = 22$; MDD: $n = 23$; SZ: $n = 22$). Groups were matched for age (mean over all groups: 40.8 ± 12.9); and sex (48% female). Education differed significantly (HC: 13.3 ± 2.0 ; MDD: 11.0 ± 1.6 ; SZ: 10.5 ± 1.8 ; $p < 0.001$). Mean MADRS (MDD) averaged 17.6 ± 7.0 , and PANSS (SZ) 63.0 ± 17.9 .

Individuals with SZ showed significantly lower SB-C scores than HC across all domains: composite ($\eta^2 = 0.16$, $p = 0.001$), memory ($\eta^2 = 0.11$, $p < 0.01$), executive functioning ($\eta^2 = 0.18$, $p = 0.001$), and processing speed ($\eta^2 = 0.14$, $p < 0.01$). Similar patterns were found in SZ vs. MDD comparisons for the composite score ($\eta^2 = 0.15$, $p < 0.01$), memory ($\eta^2 = 0.09$, $p < 0.02$), executive function ($\eta^2 = 0.18$, $p = 0.001$), and processing speed ($\eta^2 = 0.10$, $p < 0.02$). No significant differences were observed between MDD and HC in any domain ($p = 0.93$, respectively). Classification performance was highest for SZ vs. HC, with an AUC of 0.95 using an RF model based on raw RAVLT and SVF scores and speech features, compared to 0.89 with raw test scores alone based on a LM. In this comparison, features reflecting learning slope and verbal recall dynamics were most predictive. For SZ vs. MDD, classification improved with speech features, with an RF model reaching AUC = 0.85 compared to 0.82 for the LM using raw scores alone; lexical and temporal variables contributed most. In contrast, classification between MDD and HC performed worse with the additional speech features (DT, AUC = 0.54) than with raw task scores alone (LM, AUC = 0.66), where demographic variables and recall patterns played a strong role.

Conclusions: Integrating speech-derived features into standard verbal cognitive tasks enhances cognitive profiling and classification in schizophrenia, capturing disruptions in learning dynamics, timing, and lexical-semantic processing that are not visible through raw scores alone. These speech-informed metrics provide a novel, ecologically valid layer of analysis that may better reflect real-world cognitive functioning. Cognitive-linguistic features did not reliably distinguish MDD from healthy controls, potentially reflecting subtler cognitive deficits, greater individual variability, or overall lower symptom severity in the MDD group compared to the SZ cohort. However, the approach shows strong potential for differential diagnosis between SZ and MDD, and for accurate classification of SZ relative to healthy controls. These findings support the development of data-driven, low-burden digital biomarkers that align with the future of scalable, personalized, and AI-assisted assessment in precision psychiatry.

Keywords: machine learning, Cognitive impairment associated with schizophrenia, Automated natural speech analysis, cognitive assessment, precision psychiatry

Disclosure: kielelements GmbH, Employee, Self, WorldWide Clinical Trials, Consultant, Self, SanaClis, Consultant, Self.

P46. EVALUATING THE PERFORMANCE OF SIMULATED REASONING AND SELF-VERIFICATION IN LARGE LANGUAGE MODELS FOR PSYCHIATRIC CASE DIAGNOSIS

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Background: Large language models (LLMs) are being explored for applications in psychiatry, such as automatic diagnostic and therapeutic tools, due to their remarkable ability to process human language. However, these models carry significant risks, including the potential to generate inaccurate or harmful information, and prior explorations of their use in psychiatric diagnosis have noted significant overdiagnosis rates. Two emerging methods for addressing these limitations include simulated reasoning (SR) and self-verification (SV), which aim to improve model accuracy through the use of additional “reasoning” either generated by the model or provided by users. Here, we sought to evaluate the psychiatric diagnostic performance of models using SR and SV.

Methods: We extracted 106 case vignettes and associated diagnoses from the DSM-5-TR Clinical Cases book to use as our dataset. We selected the latest-generation available LLM with SR capabilities from two major vendors: o3-pro (OpenAI) and Gemini 2.5 Pro (Google). Default settings were used, with an output token limit of 2000. o3-pro was set to “medium” reasoning effort, and Gemini 2.5 Pro was set to a maximum of 16,000 reasoning tokens; safety settings were set to the minimum available value.

To test both SR and SV, we adopted two inference approaches. In the “Basic” approach, we prompted an LLM with SR directly to provide candidate diagnoses based on the case vignette. In the “SV” approach, we additionally re-prompted the model with each pairwise combination of candidate diagnoses and a query to select the most relevant diagnoses.

All resulting diagnoses from both approaches were simplified and matched to DSM-5-TR diagnoses, if possible, using a previously demonstrated semi-automated process. In this approach, modifiers were dropped, neurocognitive diagnoses were collapsed into Neurocognitive Disorder or Delirium (rather than the disease-specific disorders in the DSM), and fuzzy string matching was used to match the resulting diagnosis strings to DSM-5-TR diagnoses. Non-matching diagnoses were manually reconciled using a standardized process.

Finally, all diagnoses were scored, and the sensitivity and positive predictive value (PPV) were computed on a per-vignette basis and then averaged across all diagnoses (i.e., macro-averaged). For each model, paired t-tests were used to compare both metrics with and without SV. The macro-F1 was then computed for each model and inference approach.

Results: All inputs were successfully processed by the study LLMs, using both inference approaches. For Gemini 2.5 Pro, the sensitivity was 79.3% (SD 35.6%) and the PPV was 62.5% (SD 35.2%) without SV (F1 0.699); these values were 77.0% (SD 36.5%) and 67.3% (SD 36.4%), respectively, with SV (F1 0.718). For o3-pro, the sensitivity was 81.7% (SD 35.2%) and the PPV was 75.9% (36.4%) without SV (F1 0.787); these values were 78.2% (SD 36.1%) and 77.9% (SD 37.1%), respectively, with SV (F1 0.780). Statistical testing found a significant increase in PPV and decrease in

sensitivity for Gemini 2.5 Pro; for o3-pro, only a significant decrease in sensitivity was found.

Conclusions: We sought to evaluate the performance of two approaches to improving the accuracy of LLMs for psychiatric diagnosis using reasoning: simulated reasoning and self-verification. Overall, we found that simulated reasoning alone yielded superior predictive capabilities to previously evaluated non-SR models, with o3-pro yielding an F1 of 0.787. This represents a significant increase from the previously reported performance of gpt-4o, which yielded an F1 of 0.525 in the Basic approach. The addition of SV did not clearly improve the performance of the SR models, with mixed impacts on both. This may represent overlap or conflict between the automated reasoning token generation in the SR approach and the fixed reasoning prompt in the SV approach, and additional work is required to elucidate how best to boost the performance of SR models with prompt-based approaches.

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Keywords: Generative Artificial Intelligence, psychiatric diagnosis, Large Language Models, simulated reasoning

Disclosure: Pfizer, Stock/Equity - Publicly Traded Company, Self, P and G, Stock/Equity - Publicly Traded Company, Self, Solventum, Stock/Equity - Publicly Traded Company, Self, Viatris, Stock/Equity - Publicly Traded Company, Self, Abbott, Stock/Equity - Publicly Traded Company, Self, OpenEvidence, Stock/Equity - Privately Held Company, Self.

P47. GROWING PAINS: ASSESSING LARGE LANGUAGE MODEL PERFORMANCE ACROSS ADULT AND PEDIATRIC PSYCHIATRY

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Background: Large language models (LLMs), such as GPT-4o, are increasingly queried by patients and providers within psychiatry. Despite rapid adoption, little is known about how LLM performance (i.e., accuracy and consistency) varies across psychiatric subspecialties. Past research has suggested strong performance of LLMs on general adult psychiatry assessments; however, far less is known about LLM accuracy in content related to child/adolescent psychiatry, where diagnostic frameworks, treatment approaches, and developmental considerations can differ substantially from adult practice. Understanding potential performance gaps and deficits is essential to inform interpretation of current model results and to guide future model improvement and development. Consistent with these goals, the present study compared GPT-4o’s performance on standardized multiple-choice questions (MCQs) in adult psychiatry and child/adolescent psychiatry. We aim to highlight potential differences, identify opportunities for model improvement, and better understand LLM’s appropriateness for current use in psychiatry subspecialties.

Methods: We extracted two, 150-item psychiatry MCQ practice tests: one in adult psychiatry from Psychiatry Test Preparation and Review Manual and one in child/adolescent psychiatry from an online question bank. Using an automated, LLM framework, MCQs were classified as child-specific ($n = 139$) or non-child-specific ($n = 161$) based on case age, diagnosis, developmental context, and treatment type. Manual review of 50 randomly selected MCQs (25 in each category) found no errors in this automated classification.

Using best-practice prompting approaches, including persona and instruction-based prompting, GPT-4o was prompted to independently answer every MCQ 50 times. We calculated accuracy (% correct), pooled accuracy (% correct, using the most common MCQ response across trials, i.e. for three trials, responses 'B, A, A' would become 'A'), and response consistency (how often the model selected the same answer for a given question). T-tests compared results across test-type (adult vs. child) and question category (child-specific vs. non-child-specific). Low-accuracy questions were qualitatively reviewed to elucidate themes and error patterns.

Results: GPT-4o demonstrated lower accuracy on child (71.1%) vs. adult (86.9%) practice tests, $p < 0.001$. Pooled analysis showed that the accuracy gap between child and adult practice tests remained statistically significant when answers were combined across up to 17 trials (i.e., using the most frequent MCQ response across repeated trials; $p < 0.05$), but disappeared once 18 trials were combined. Response consistency was also lower for the child practice test ($p < 0.001$). GPT-4o's accuracy gap was wider among child-specific questions (68.1%) vs. non-child-specific (88.4%), $p < 0.001$, and consistency was lower, $p < 0.001$. Pooled analysis showed that the accuracy gap remained statistically significant when responses were combined across up to 18 combined trials ($p < 0.05$), but disappeared at 19. Manual review of low-accuracy MCQs highlighted recurring errors in developmental disorders, pediatric medication use, and early childhood presentations (ages 2–6). GPT-4o seemed to bias towards adult psychiatric practice (e.g., erroneously overlooking guanfacine as a treatment for ADHD) or disorders that are more prevalent and likely appear with higher frequency on the internet (e.g., erroneously selecting autism spectrum disorder over global developmental delay).

Conclusions: GPT-4o shows significantly lower accuracy and consistency in child/adolescent psychiatry vs. adult MCQs. Observed errors in our analysis appeared more frequent for content related to developmental disorders, pediatric medication use, and early childhood presentations, with many responses suggesting model bias towards high-frequency terms and an adult psychiatry knowledge base. Pooled analysis suggests that when given enough chances to answer MCQs (i.e., 18 or 19 tries per question), GPT-4o eventually performs just as well on child psychiatry as adult psychiatry, suggesting it does contain accurate pediatric knowledge — but with far more uncertainty and inconsistency in how that knowledge is accessed. While the training datasets for GPT models are not publicly available, this finding suggests that GPT-4o may have been trained on limited or unreliable child and adolescent psychiatry content, possibly including online misinformation. These findings underscore the need for caution when applying LLMs in child and adolescent psychiatry and point to the importance of future studies that evaluate and compare model performance across other psychiatric subspecialties, examine performance in more clinically relevant scenarios, and test strategies for reducing uncertainty and bias in pediatric contexts.

Keywords: Artificial Intelligence, Analysis, Neuroscience, Clinical Neuroscience, Pharmacology, machine learning, child and adolescent psychiatry

Disclosure: Nothing to disclose.

P48. PREDICTING SUBSTANCE USE AND PSYCHOSIS OUTCOMES IN ADOLESCENCE

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Background: The profound structural and functional maturational changes in the brain during adolescence heighten susceptibility to environmental influences. Adolescence is further characterized by an increased propensity for substance use behaviors that contribute to the emergence of psychopathology. There is high comorbidity between substance use and schizophrenia in adulthood, and evidence for increased rates of substance use among adolescents displaying psychotic symptoms. Despite these associations, the link between substance use, psychosis, and their associated risk factors remain poorly understood. There is a need to elucidate how the combination of these multifaceted risk factors, including demographic, clinical, and neural factors collectively influence individual risk during adolescent development. Applying machine learning classification models on large, longitudinal data holds promise for improving our prediction of negative health outcomes.

Here, we leverage data from the Adolescent Brain and Cognitive Development (ABCD) Study, aiming to predict risk for substance use, psychosis spectrum, and comorbid outcomes in adolescence from childhood risk factors via machine learning classifiers. We hypothesized that shared and distinct demographic, clinical, and neuroimaging features would differentially predict early psychotic-like experiences (PLEs), substance use endorsement (SU), and co-occurrence of PLEs and SU, reflecting partially overlapping but dissociable risk profiles.

Methods: ElasticNet machine learning models were trained on baseline ABCD data ($N = 4,735$; ages 9–10; 44.9% boys) to predict SU defined by cannabis, alcohol, or tobacco use endorsement and PLEs as measured by the Prodromal Questionnaire–Brief Child Version in adolescent boys and girls (timepoint 4; ages 13–14). Outcomes were categorized into four groups: (1) PLE, (2) SU, (3) comorbid PLE + SU, and (4) neither outcome. Baseline predictors included demographic and clinical variables (e.g., sex, income, trauma, family history), and multimodal neuroimaging features: task-based fMRI (e.g. activation of the striatum and ventromedial prefrontal cortex to reward anticipation from the Monetary Incentive Delay (MID) Task, activation of the amygdala and hippocampus to negative faces from the Emotional nBack (eNBack), prefrontal cortex activation during failed inhibition from the Stop Signal Task (SST)), structural MRI, resting-state functional connectivity (rsFC), and diffusion imaging. Model performance was evaluated with ROC curves (AUROC); feature importance was assessed using coefficient weights.

Results: Models incorporating integrated sets of neural, clinical, and demographic features from baseline data were strong predictors of PLE (AUC = 0.94) and SU (AUC = 0.72) outcomes in adolescence. Demographic and clinical factors such as internalizing symptoms, parental drug and alcohol use, parental depression, and lower family income showed the strongest predictive effects, while neural features provided additional predictive value and improved model accuracy. Some predictors, such as parental depression and lower socioeconomic status, contributed to both outcomes, while others were outcome-specific. Specifically, internalizing symptoms were the strongest predictor of PLEs ($\beta = 0.021$), whereas parental drug use was most predictive of SU ($\beta = 0.22$).

Key neural predictors of PLEs included cingulo-opercular network (CON; $\beta = -0.01$), hypoactivation of the amygdala ($\beta = -0.006$), prefrontal thickness ($\beta = -0.003$), inferior frontal gyrus during failed task inhibition (SST; $\beta = 0.003$), and amygdala hypoactivation in response to negative versus neutral faces (eNBack; $\beta = -0.006$).

In contrast, key neural predictors of SU included amygdala activation ($\beta = 0.20$), hippocampal activation ($\beta = .10$), and fusiform activation ($\beta = 0.12$) to negative versus neutral faces; striatal ($\beta = -0.01$) and ventromedial prefrontal cortex activation ($\beta = 0.04$) to the anticipation of reward (MID); CON rsFC ($\beta = -0.04$) and default mode network (DMN) rsFC ($\beta = -0.005$); and increased transverse diffusivity of the pars triangularis ($\beta = 0.06$). Thus, while both models implicated amygdala/hippocampal reactivity and CON rsFC, PLEs were uniquely associated with internalizing symptoms and inhibitory control deficits, whereas SU was more strongly linked to familial SU history, reward-related activation, and fusiform reactivity.

Co-occurrence was driven by a distinct risk profile (AUC = 0.82), characterized by effects of parental drug use ($\beta = 0.65$), internalizing symptoms ($\beta = 0.45$), parental depression ($\beta = 0.20$), and trauma ($\beta = 0.10$), with neural contributions that overlapped with PLEs (amygdala $\beta = 0.12$ and hippocampal $\beta = 0.42$ reactivity; CON rsFC $\beta = -0.35$) and SU (fusiform activation $\beta = 0.13$; striatal reward anticipation $\beta = 0.05$; DMN rsFC $\beta = -0.18$), as well as effects of white-matter integrity (forceps minor diffusivity; $\beta = 0.20$).

Conclusions: This study provides support for shared and distinct predictors of SU and psychosis outcomes in adolescence. Predictive models integrating demographic, clinical, and neural variables achieved high performance. Some features contributed similarly, while others showed outcome-specific effects. Results highlight that comorbidity is not simply additive but reflects a distinct risk signature. Our results identify early indicators of these conditions in ways that may inform treatment strategies.

Keywords: Psychosis-Risk, Substance Use Initiation, Schizophrenia (SCZ), machine learning classification, Adolescence

Disclosure: Nothing to disclose.

P49. UNCOVERING CAUSAL PATHWAYS LINKING PARENTAL BEHAVIORS, GENETIC RISK, AND EXTERNALIZING BEHAVIORS IN CHILDREN USING A DATA-DRIVEN FRAMEWORK

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Background: Externalizing behaviors in children, such as aggression, hyperactivity, and defiance, arise from complex interplays between genetic predispositions and environmental influences, particularly parental behaviors. Clarifying these intricate causal pathways can benefit from data-driven discovery approaches.

Methods: We developed Hillclimb-Causal Inference, a causal discovery framework that combines the Hill Climb Search algorithm with a customized Linear Gaussian Bayesian Information Criterion (BIC). Using data from the Adolescent Brain Cognitive Development (ABCD) Study, we analyzed parental behavior assessments, children's genotypes, and measures of externalizing behaviors. Dimensionality reduction was applied to address multicollinearity among parental behaviors. Children's genetic liability was quantified using polygenic risk scores (PRS) for externalizing disorders. Identified pathways were validated and quantified using structural equation modeling (SEM).

Results: Three primary causal patterns emerged. (1) Parental alcohol, tobacco, and substance-use were interrelated, influenced one another, and jointly contributed to broader parental

behavioral problems. (2) Among parental influences, alcohol misuse exerted the strongest direct effect on children's externalizing behaviors ($\beta = 0.33$), followed by broader externalizing/internalizing problems reported by spouses ($\beta = 0.20$). In contrast, self-reported parental problems showed weaker effects, suggesting potential biases in self-assessments compared to external observations. (3) Children's PRS contributed modestly to externalizing outcomes ($\beta = 0.07$) but remained significant, highlighting a genetic component. Collectively, when considering both direct and indirect paths, parental substance misuse (alcohol, drug, tobacco) yielded a total effect exceeding 1.1 on externalizing behaviors. Bootstrap resampling and sensitivity analyses confirmed the robustness of these pathways.

Conclusions: Hillclimb-Causal Inference enabled robust identification of causal pathways, supported by SEM, resampling, and sensitivity checks. Our findings underscore the critical influence of parental alcohol and substance use, alongside broader externalizing and internalizing behavioral problems, in shaping children's externalizing outcomes, while also highlighting a smaller yet meaningful genetic risk as reflected in PRS. These results suggest that interventions targeting parental substance use and related behaviors may be particularly effective in reducing externalizing problems during adolescence and buffering the genetic risk. This work illustrates the utility of data-driven causal inference as a general framework for studying complex causal mechanisms in psychiatric genetics.

Keywords: Causal Pathways, Externalizing Behavior, Parental Factors, Genetic Risk, Adolescence

Disclosure: Nothing to disclose.

P50. METRIC LEARNING-ENHANCED PHENOTYPIC SCREENING IN ZEBRAFISH FOR IDENTIFICATION OF GABAERGIC ANALGESICS AND ANTIDOTES

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Background: Psychiatric drug discovery remains challenging because traditional target-based approaches fail to capture the complexity of brain circuits and behavior, while phenotypic screening generates rich but difficult-to-interpret behavioral datasets at a large scale. Here, we tested whether deep metric learning can accelerate behavioral phenotyping and uncover structure-activity relationships (SAR) in a compound library of sedative hypnotics on zebrafish.

Methods: We trained a supervised twin neural network on motion index time-series from larval zebrafish exposed to a high-replicate dataset of 650 FDA-approved reference compounds, optimized with a contrastive loss function to learn embeddings that preserve behavioral similarity in drug replicates. The model was applied to ~300 pyrazoloquinolinones (PQs), a chemically diverse class of GABA-A receptor modulators with complex pharmacology.

Results: Using the embeddings as input for a density-based spatial clustering (DBSCAN) algorithm, compounds self-organized into functional categories: GABA-A agonists cluster with the benzodiazepine control midazolam, competitive antagonists form a distinct cluster, and inactive compounds group with vehicle (DMSO). Notably, clustering revealed SAR within the PQ scaffold, identifying substituents enriched in the different phenotypic clusters. Misclassification of inverse agonists as inactive compounds highlighted a current limitation, as their stimulant-like phenotypes produced stochastic, heterogeneous locomotor signatures.

Conclusions: These results demonstrate that machine learning-aided zebrafish behavioral profiling provides a framework for discovering behavioral structure-activity-relationships in psychoactive compounds. This approach illustrates a path to reviving phenotypic screening augmented with artificial intelligence in therapeutic domains, such as neuropsychiatric disorders, where molecular mechanisms remain elusive.

Keywords: Deep learning, Behavioral Phenotyping, Drug Discovery - new approaches, zebrafish, GABAA receptor positive allosteric modulator

Disclosure: Nothing to disclose.

P51. ALTERED WHITE MATTER DEVELOPMENT AFTER CONCUSSION LINKED TO ANXIOUS BEHAVIORS IN CHILDREN AND MICE

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Background: Myelin is a lipid-rich, multilaminar membrane synthesized by oligodendrocytes glia cells in the brain that ensheathes axons to facilitate rapid saltatory conduction of action potentials and provide metabolic support. Babies are born with very little myelin and myelination of axons by oligodendrocytes continues into early adulthood influencing circuit maturation and function. Mild traumatic brain injuries (mTBI, aka concussions) are common in children and some children experience long-lasting emotional and behavioral problems. Our recent studies have identified premature appearing white matter microstructure in female children with a history of concussion and enhanced emotional problems. This presentation will describe altered longitudinal development of white matter in female children with prior concussion and the potential cellular underpinning in a juvenile mouse model of mTBI that results in enhanced anxiety-like behaviour.

Methods: To assess longitudinal development of white matter following concussion early in life we examined children from the Adolescent Brain Cognitive Development Study that experienced a concussion before study entry at 9–10 years old and had baseline and 2 year follow up MRI data (concussion N = 202, comparison group N = 4537). Change in directionally restricted diffusion from the Restriction Spectrum Imaging model fit to the multi-shell diffusion-weighted MRI data was assessed as a measure of axon density development in deep and superficial white matter. To determine potential cellular drivers of altered white matter maturation after concussion we established a juvenile mouse model of mTBI that results in elevated anxiety like-behaviours. Mice underwent closed-skull impacts centered on bregma once daily from postnatal days 17 to 19. Mice were sacrificed one or four weeks post-injury for brain analyses. Immunohistochemistry was conducted to quantify the density of total (CC1) and newly generated oligodendrocyte progenitor cells (pgcrlalpha+EdU) and oligodendrocytes (CC1 +EdU). Behavioral assessments (elevated zero maze, and light-dark box) were performed to evaluate emotional and cognitive impairments.

Results: Trajectories of deep white matter development were altered in a sex dependent manner by concussion history before age 9–10. Females with prior concussion demonstrated less age dependent maturation and males demonstrated more age dependent maturation compared to children with no history of mTBI (linear mixed effects model: group x time x sex interaction $\chi^2 = 2.4 \times 10^{-4}$, $p = 0.059$). In the juvenile mouse model there was a greater number of newborn oligodendrocytes (CC1 +EdU) in the

corpus callosum one week, but not four weeks, after mTBIs compared to sham mice (group-by-time interaction, $F(1,11) = 7.359$, $p = 0.020$, partial eta squared $\eta^2 = 0.40$). Additionally, one week post-mTBI, mice spent less time in the open arms of the elevated zero maze ($t = 2.074$, $p = 0.049$, Cohen's $d = -0.766$), and had a higher latency to enter the light area in the light-dark box test ($t = -2.178$, $p = 0.033$, Cohen's $d = 0.95$) compared to sham-injured mice.

Conclusions: The observed increase in oligodendrogenesis in the corpus callosum of juvenile mice after mTBI suggests that this may play a role in the abnormal white matter development seen in children, which is linked to anxiety problems.

Keywords: anxiety, concussion, oligodendrocytes, myelin, white matter

Disclosure: Nothing to disclose.

P52. MEASURING BRAIN FATTY ACID AMIDE HYDROLASE LEVELS IN INDIVIDUALS WITH SOCIAL ANXIETY DISORDER USING THE PET TRACER [C-11]CURB

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Background: Social Anxiety Disorder (SAD) impacts up to 12% of individuals, with females reporting higher prevalence and greater symptom severity. Response to current treatments remains low, necessitating new treatment strategies. Fatty acid amide hydrolase (FAAH), the enzyme that metabolizes the endocannabinoid anandamide (AEA), has emerged as treatment target for SAD. Preclinical investigations have reported a link between stress exposure, increased FAAH, and reduced AEA. Elevating AEA levels by inhibiting FAAH decreases anxious behavior and enhances social interaction in rodents. A recent human clinical trial in SAD reported a > 30% reduction in the Lebowitz Social Anxiety Scale (LSAS) measurement of symptom severity after FAAH inhibition, which was associated with greatest change in peripheral AEA. No research to-date has investigated brain FAAH in individuals with SAD.

The positron emission tomography (PET) tracer [11C]CURB enables direct quantification of brain FAAH levels in humans. This study aims to use PET imaging of [11C]CURB to test the hypothesis that brain FAAH levels are higher in SAD relative to healthy controls (HCs) specifically selected for low social anxiety. A secondary, preliminary aim is to investigate if higher FAAH is associated with elevated SAD symptom severity according to the LSAS.

Methods: Individuals meeting DSM-5 criteria for SAD and scoring > 60 on the LSAS were invited as part of the SAD group. Individuals without a history of psychiatric diagnosis and scoring below 30 on the LSAS participated as Healthy Controls (HCs). Participants completed a PET scan with the FAAH probe, [11C]CURB, a magnetic resonance imaging scan, the LSAS, and clinical assessments and questionnaires. Blood samples were taken to assess genetic variability of the FAAH (C385A) polymorphism known to contribute to its variability. In-house software was used to delineate regions of interest (ROI), extract time activity curves and complete kinetic modeling, producing a reliable index of FAAH levels, λ_{k3} . A repeated measures (RM) ANCOVA was used to assess FAAH differences with diagnostic group (SAD, HC) and sex (male, female) as between-subject factors, and ROI as the within-subject factor. FAAH genotype was included as a covariate. The relationship between whole brain FAAH (average of 9 ROIs) and

LSAS total score in participants with SAD was assessed with partial correlations controlling for FAAH genotype.

Results: Thirty-one individuals with SAD (M/F: 10/21; 26.32 ± 5.9 years old) and 22 HCs (M/F: 10/12 25.09 ± 4.8 years old) completed the study. There were no differences in demographic variables across groups (sex, age, BMI, FAAH genotype; $p > 0.05$). The RM ANCOVA revealed a trend for lower whole brain FAAH in individuals with SAD by 6%, but this did not reach statistical significance ($F(1,48) = 3.25$, $p = 0.077$, $\eta^2_p = 0.06$). The diagnostic group-by-ROI interaction was not statistically significant ($F(3.15,151.15) = 1.94$, $p = 0.120$), but individuals with SAD had significantly lower FAAH in the hippocampus (-8% , $p = 0.041$), temporal cortex (-8% , $p = 0.027$), and thalamus (-9% , $p = 0.028$). The analysis did not reveal a main effect of sex ($F(1,48) = 2.02$, $p = 0.162$, $\eta^2_p = 0.040$). Furthermore, the analysis did not find a significant diagnostic group-by-sex interaction ($F(1,48) = 0.47$, $p = 0.486$, $\eta^2_p = 0.010$), although females with SAD demonstrated a non-significant trend for lower brain FAAH compared to female HCs by 8% ($p = 0.053$), while there were no differences among male groups ($p = 0.468$).

The partial correlation assessing the relationship between LSAS total scores and whole brain FAAH revealed no significant correlation in the entire SAD group ($n = 31$, $r = 0.18$, $p = 0.33$). Sex-disaggregated analysis showed a significant positive correlation in females ($n = 21$, $r = 0.48$, $p = 0.031$), but a negative trend was observed in males ($n = 10$, $r = -0.39$, $p = 0.31$).

Conclusions: This first in human study offers insight into brain FAAH levels in SAD and the relationship with social anxiety symptom severity. A trend for lower whole brain FAAH in SAD compared to HCs was reported. Although the group-by-sex interaction was not significant, another trend for lower FAAH was reported in females with SAD, but not males with SAD compared to sex-matched HCs. Brain FAAH was positively correlated with SAD symptom severity in females, suggesting a potential relationship between higher FAAH and higher symptom severity. The finding of a trend for lower FAAH in SAD does not support the direction of the initial hypothesis of higher FAAH in SAD based on preclinical literature. The analysis highlights the importance of considering sex differences in FAAH-related research as results may differ between males and females. Overall, these findings may suggest that FAAH levels in the absence of acute social stress may be lower or may not differ in individuals with SAD compared to HC, while higher FAAH may be positively associated with higher symptom severity, specifically in females. This preliminary observation may suggest that FAAH levels in the human brain may be linked to state rather than trait levels of social anxiety. More research is needed to explore how chronic and acute social stress exposure affects FAAH and endocannabinoid signalling in support of the investigation of endocannabinoid targeting treatments.

Keywords: PET Imaging Study, Social Anxiety Disorder, Endocannabinoid system, Fatty Acid Amide Hydrolase

Disclosure: Nothing to disclose.

P53. TREATMENT-RELATED CHANGES IN FUNCTIONAL ACTIVATION DURING A THREAT-PROCESSING TASK AMONG YOUTH WITH ANXIETY DISORDERS: AN INDEPENDENT REPLICATION

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Background: Anxiety disorders are among the most common and impairing psychiatric conditions in youth. Cognitive-behavioral

therapy (CBT) is an effective first-line non-pharmacological treatment. Previous research (Haller et al., 2024) has demonstrated that activation in fronto-parietal networks may normalize following CBT for youth with anxiety disorders. The current study investigates whether these effects indeed replicate in a large, independent sample of youth.

Methods: An independent sample of pediatric patients ages 8–18 with a primary anxiety diagnosis ($n = 67$) underwent 12 weeks of CBT as a part of two randomized controlled trials (White et al., 2017; Linke et al., 2019; Byrne et al., in press). Patients completed a threat-processing dot-probe task during functional MRI before and after treatment. An age-matched independent sample of healthy comparison youth ($n = 90$) completed the same task during two scans over the same time interval. Whole-brain regional activation changes (thresholded at $p < 0.001$) were examined using task-based blood-oxygen-level-dependent (BOLD) response. Analyses included age, sex, and cohort as covariates.

Results: Significant ($p < 0.001$) group-by-time interactions were observed for 38 clusters, with notable similarities between several clusters and previous research. Post-hoc tests revealed a potential treatment-induced compensatory effect in the right inferior parietal lobule, replicating past work of Haller et al. 2024. Namely, there were no pretreatment differences between the anxiety and control groups in the right inferior parietal lobule, whereas the anxiety group showed significantly less activation compared to the control group at the posttreatment scan. This pattern was also observed in the right amygdala in the opposite direction. While there were no pretreatment differences between groups, youth with anxiety exhibited increases in right amygdala activation after treatment compared to healthy controls, which replicates past findings of Maslowsky et al. 2010. Lastly, a notable cluster in the left middle cingulate cortex demonstrated reduced pre-treatment activation among the anxiety group compared to healthy controls. Posttreatment activation in this region increased among the anxiety group, with no change in the healthy comparison group, similar to past findings of Burkhouse et al. 2018.

Conclusions: Findings reveal a partial but inconsistent replication of past work on neural changes following the acute effects of CBT for pediatric anxiety. While several key clusters demonstrated similarities with prior research, there was no evidence of fronto-parietal normalization following CBT for an independent sample of youth with anxiety disorders. Mixed findings highlight the need for integrating past findings using a Bayesian approach in future work. Identifying potential cortical and subcortical targets before and after treatment may help to refine clinical approaches for pediatric anxiety.

References:

Burkhouse, Katie L., et al. "Anterior cingulate activation to implicit threat before and after treatment for pediatric anxiety disorders." *Progress in Neuro-Psychopharmacology and Biological Psychiatry* 84 (2018): 250–256.

Byrne, Meghan E. et al. "Gamified Attention Bias Modification Training to Augment CBT for Youth Anxiety Disorders: A Randomized Controlled Trial." *Journal of the American Academy of Child and Adolescent Psychiatry* (in press).

Haller, Simone P. et al. "Normalization of fronto-parietal activation by cognitive-behavioral therapy in unmedicated pediatric patients with anxiety disorders." *American Journal of Psychiatry* 181.3 (2024): 201–212.

Linke, Julia O. et al. "Efficacy and mechanisms underlying a gamified attention bias modification training in anxious youth: protocol for a randomized controlled trial." *BMC Psychiatry* 19.1 (2019): 246.

Maslowsky, Julie, et al. "A preliminary investigation of neural correlates of treatment in adolescents with generalized anxiety

disorder." *Journal of Child and Adolescent Psychopharmacology* 20.2 (2010): 105-111.

White, Lauren K. et al. "Complementary features of attention bias modification therapy and cognitive-behavioral therapy in pediatric anxiety disorders." *American Journal of Psychiatry* 174.8 (2017): 775-784.

Keywords: Children and Adolescents, Functional MRI (fMRI), Replication, Pediatric anxiety disorders, Anxiety-related circuitry

Disclosure: Nothing to disclose.

P54. JOINT EFFECTS OF CORTISOL RECOVERY AND THREAT-RELATED ATTENTIONAL BIAS IN PREDICTING ANXIETY SYMPTOMS IN HEALTHY CHILDREN: A LONGITUDINAL STUDY

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Background: Anxiety disorders are prevalent in children and significantly impair their functioning. Their prevalence increases during adolescence, especially in girls. The brain expresses cortisol receptors throughout life, and during development it is especially sensitive to cortisol dysregulation. Dysregulated cortisol secretion is thought to contribute to the emergence and maintenance of clinical anxiety. Consistent with this view, we recently showed that poorer cortisol recovery after a psychosocial stressor prospectively predicted steeper growth in anxiety symptoms in healthy children, especially in girls. This highlights cortisol recovery as a candidate endocrine risk marker.

Cognitive patterns associated with anxiety may amplify this vulnerability. Laboratory studies consistently show selective attention to threat in anxious youth: they detect angry faces more quickly (facilitation) and have difficulty disengaging from them. In our prior work, we also observed similar anger-related biases among healthy children with a socio-emotional vulnerability profile. Because cortisol receptors are abundant in neural circuits that regulate threat perception, alterations in cortisol recovery may shape how children detect and disengage from threat. Accordingly, cognitive patterns linked to anxiety—such as selective attention to anger—may compound this cortisol-related vulnerability.

Building on this foundation, we tested whether anger-related attentional bias and cortisol recovery act additively to shape the longitudinal course of anxiety symptoms in healthy children. We hypothesized a moderation pattern: the link between cortisol recovery and later anxiety would be stronger at higher levels of anger-related attentional bias, alongside independent main effects of each risk factor.

Methods: We conducted a longitudinal study in healthy children ($N = 111$; ages 8–12 [$M = 10.14$] at T1). Anxiety symptoms were assessed at baseline (T1) and re-assessed 6 (T2) and 18 months (T3) later using the Behavior Assessment System for Children, Third Edition. At T1, children completed the Trier Social Stress Test for Children (TSST-C); seven saliva samples were collected for cortisol quantification. From these samples we derived two physiological stress indices. Cortisol reactivity was indexed by AUCi (area under the curve with respect to increase), computed via the trapezoidal method relative to baseline. Cortisol recovery was quantified as the percent decline from peak to the final sample.

Threat-related attentional bias was measured with a computerized Visual Search Task with 3×3 arrays of faces or objects. Two conditions indexed complementary processes: facilitation to anger (locate a threat target among eight neutral distractors) and disengagement from anger (locate a neutral target among eight threat distractors). Reaction time (RT; ms) was the outcome after

standard RT cleaning: incorrect trials were removed; anticipations and time-outs were excluded ($RT < 200$ ms or > 3000 ms); and, within participant and condition, RTs beyond ± 3 SD of the mean were trimmed. Cleaned RTs were then averaged to yield the facilitation-to-anger and disengagement-from-anger indices (higher RT = slower processing).

To account for established risk factors, we created a socio-emotional vulnerability composite by averaging z-scores from anxiety sensitivity, intolerance of uncertainty, and perseverative cognition scales; additional variables were pubertal status and parental history of an internalizing disorder.

We tested whether attentional bias and cortisol recovery exert interactive effects on the longitudinal trajectory of anxiety using linear mixed-effects models with time (T1–T3) and the above predictors, adjusting for covariates. Analyses were run in Python (statsmodels). Motivated by prior work showing cortisol-recovery effects in girls, all models were sex-stratified.

Results: In boys, socio-emotional vulnerability uniquely predicted higher anxiety across time ($\beta = 0.59$, $p = 0.0061$, 95% CI [0.34, 0.85]; $R^2 = 0.42$). In girls, there was a significant cortisol recovery $\times$ facilitation-to-anger $\times$ time (3 levels) interaction on anxiety symptoms. Post-hoc tests indicated that the T3-specific interaction term was significant ($B = -2.41 \times 10^{-5}$, $p = .0037$, 95% CI $[-4.03 \times 10^{-5}, -7.84 \times 10^{-6}]$). Simple slopes at T3 showed that with high facilitation-to-anger (+1 SD; slower reaction times) the association between cortisol recovery and anxiety was not significant ($B = 4.3 \times 10^{-5}$, $p = .874$, 95% CI $[-4.83 \times 10^{-4}, 5.68 \times 10^{-4}]$). At low facilitation-to-anger (−1 SD; faster reaction times) the slope was positive and significant ($\beta = 0.89$, $p = .00086$, 95% CI $[-1.41, -0.37]$). In other words, among girls with faster reaction times on facilitation-to-anger trials, we observed a positive association between the cortisol-recovery measure (reflecting poorer recovery) and anxiety symptoms. The full girls' model explained $R^2 = 0.661$, and the three-way interaction accounted for $R^2 = 0.140$.

Conclusions: While girls' anxiety trajectories were jointly shaped by cortisol recovery and threat-related attentional bias, boys' anxiety symptoms were best predicted by self-reported socio-emotional vulnerability. These results clarify sex-specific mechanisms in youth anxiety development, providing a rationale for mechanism-based interventions.

Keywords: Cortisol response to stress, Anxiety development, youth, attentional bias

Disclosure: Nothing to disclose.

P55. THE ASSOCIATION BETWEEN OBSESSIVE-COMPULSIVE PERSONALITY DISORDER TRAITS AND HEALTH-RELATED QUALITY OF LIFE IN INDIVIDUALS WITH ANXIETY AND MOOD DISORDERS

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Background: Impaired Health related Quality of Life (HRQoL) is a common burden among individuals with mental health conditions, especially in those with anxiety and mood disorders (AMD). Obsessive-compulsive personality disorder (OCPD) is one of the most common personality disorders in the general population, is even more prevalent in individuals with AMD, and has also been linked to excessive mental fatigue in this group. Nevertheless, there have been very few studies exploring the impact of OCPD on HRQoL in patients with AMD.

Our objective was to investigate the relationship between OCPD and HRQoL in individuals with AMD.

Methods: A cross-sectional study included 235 persons (with a mean age of 41 and SD of 12 years) with AMD attending the Stress Disorders Unit at Hospital Palanga Clinic, Neuroscience Institute, Lithuanian University of Health Sciences. The SF-36 scale was used to assess eight aspects of HRQoL: physical functioning, role-physical functioning, bodily pain, general health, vitality, social functioning, role-emotional functioning, and mental health. The Compulsive Personality Assessment Scale (CPAS) was used to assess features associated with OCPD. The nine-item Patient Health Questionnaire (PHQ-9) and seven-item Generalized Anxiety Disorder scale (GAD-7) were used to assess depression and anxiety symptoms in individuals with AMD.

Two-tailed Student's *t*-test or Fisher's χ^2 were used to compare socio-demographic, clinical, HRQoL, depression, and anxiety characteristics between individuals with and without OCPD. Subsequently, separate multivariable linear regression analyses were performed within each group (OCPD and non-OCPD) to examine the associations between OCPD traits and HRQoL, while controlling for potential confounders.

Results: Subjective social functioning, mental health, vitality and general health perception were found to be worse in individuals with OCPD in comparison to group with no OCPD. In individuals with OCPD, the links between OCPD traits and HRQoL remained significant only in the domain of vitality ($\beta = -0.236$, $p < 0.042$), after controlling for age, gender, education, marital status, medication use and depressive and anxiety symptoms. OCPD trait severity was not associated with HRQoL domains in individuals without OCPD.

Conclusions: In individuals with AMD, the presence of OCPD was related to poorer HRQoL, particularly in the vitality domain, after controlling for socio-demographic and clinical characteristics. This study sheds new light on the impact of OCPD on quality of life in individuals with AMD and aligns with our previous findings linking OCPD to mental fatigue. Limitations of the study include its cross-sectional design and use of brief measures to assess anxiety/depressive symptom severity. Future research should now explore mechanisms underlying these associations, and evaluate whether the findings generalise to larger, more diverse samples.

Keywords: Obsessive-compulsive personality traits, health-related quality of life, Anxiety, Depression

Disclosure: Nothing to disclose.

P56. PILOT STUDY OF A TWO-SESSION POSITIVE EMOTION FOCUSED TREATMENT FOR INTERNALIZING PSYCHOPATHOLOGY: NEUROBIOLOGICAL PREDICTION OF TREATMENT OUTCOME AND TARGET ENGAGEMENT

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Background: Positive emotion focused psychotherapies appear to be efficacious for depression and anxiety, with emerging evidence supporting brief versions of these interventions. To whom these treatments are best-suited and their underlying mechanisms are, nonetheless, poorly understood. One possibility is that positive emotion focused treatment might be best-suited to patients without exaggerated responding to negative stimuli. Further, positive emotion focused treatments are proposed to target positive emotion up-regulation deficits previously observed in internalizing psychopathology. In this preliminary study, we used EEG measure, the late positive potential (LPP) – a measure of emotion-processing – to predict treatment gain and assess target

engagement during our novel, two-session internalizing intervention, Short-term Methods for Increasing Life's Enjoyment (SMILE).

Methods: Participants who had moderate or higher levels of internalizing psychopathology were randomly assigned to two weekly sessions of SMILE or a control condition. In Study 1 ($N = 51$; $n = 24$ SMILE, $n = 27$ control), the LPP elicited by pre-treatment passive viewing of positive and negative (versus neutral) pictures was used to predict worry at post-treatment (two weeks later), controlling for baseline worry. In Study 2, ($N = 39$; $n = 20$ SMILE, $n = 19$ control) we examined treatment effects on the LPP during savoring (versus viewing) of positive and neutral pictures.

Results: Positive affect treatment reduced depression, suicidality, worry and other symptoms, $-0.89 < \beta s < -0.36$, $ps < .02$, with benefits retained at three-months for several measures. In Study 1, patients in the SMILE group with smaller LPPs to negative pictures at baseline showed greater reductions in worry from pre- to post-treatment [$t(47) = 2.29$, $p = 0.027$] with no effect observed for the control group. In Study 2, savoring of positive pictures was greater in the SMILE group relative to the control group at post-treatment, $F(1,37) = 6.26$, $p = 0.017$ (no differences were observed between groups at pre-treatment). Initial results suggested that target engagement did not mediate treatment-related change in worry.

Conclusions: Individuals with reduced electrocortical response to negative pictures at pre-treatment showed the greatest treatment-related reductions in worry. Therefore, SMILE may be best suited to patients without exaggerated response to negative stimuli. Interestingly, patients with blunted response to negative stimuli tend to do poorly in traditional, negative-emotion focused psychotherapy; as such, results suggest that SMILE may provide a useful alternative for these individuals. In keeping with an experimental therapeutics approach, SMILE also engaged a treatment target (savor > view LPP) that we have previously found to be associated with depression. These results provide a measure of the neurobiological effect of treatment, though lack of mediation suggests that savoring may not be the primary mechanism at work in SMILE. Other possibilities, such as insufficient effect size or reverse causality (e.g., changes in worry lead to changes in savoring) should be considered. Overall, results shed light on patient profiles that may be best suited to positive affect therapy and inform hypotheses for future investigations of treatment mechanism.

Keywords: Positive Affect Treatment, EEG/ERP electrophysiology, internalizing disorders, Emotional regulation, personalized medicine

Disclosure: Nothing to disclose.

P57. CHILD ANXIETY AND PARENTAL ACCOMMODATION LINKED WITH ALTERED DEVELOPMENTAL PATTERNS OF INDEPENDENT FEAR PROCESSING

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Background: Parents play a powerful, shifting role in their child's fear regulation over the course of typical development. In typical development, parental presence can buffer youth's neurobiological fear responses. Specifically, children show dampened amygdala responses to parental cues and reduced ventromedial prefrontal cortex (vmPFC) activation to conditioned threat cues when conditioning occurs in a parent's presence. These neural regions undergo dynamic structural and functional changes that

parallel developmental changes in fear regulation. Whereas healthy youth show a developmental shift from parent-dependent to independent fear responding, this developmental shift may be disrupted in children with anxiety disorders.

Parenting behaviors play an important role in childhood anxiety disorders. For example, parental presence can encourage approach-related behaviors that are important for treatment. However, parenting behaviors that encourage reliance on parents may have long-term consequences for anxious children's fear processing. The vast majority of parents of anxious children engage in parental accommodation of anxiety (i.e., changing their own behaviors and schedules to mitigate their child's anxiety). Family accommodation negatively impacts youth self-efficacy and may alter developmental trajectories of independent fear processing. In the current study, we examine how child anxiety and family accommodation relate to age-related patterns of independent fear processing.

Methods: Participants were 196 6- to 12-year-old youth who met criteria for a primary anxiety disorder ($M = 8.99$, $SD = 1.76$; 90 female, 106 male) and their parents. Child participants and their parents completed the Family Accommodation Scale for Anxiety and the Pediatric Anxiety Rating Scale. Child participants also completed two runs of an event-related fMRI task measuring parental modulation of children's neural reactivity to fearful face stimuli. In each run, participants view a series of face stimuli with fearful and neutral face stimuli. Child participants complete two runs of this task: one while alone in the scanner room ("Alone Condition") and one while their parent is physically present in the scanner room holding their child's hand ("Parent Condition"). The order of these task conditions is counterbalanced across participants. To examine anxiety- and accommodation-related changes in expected developmental trends in parent-modulated and independent fear processing, we conducted region-of-interest analyses focusing on the amygdala and vmPFC. All analyses controlled for child sex-at-birth, scan order, and study phase (data were collected as part of a two-phase R61/R33-funded study). Analyses related to child anxiety controlled for parental accommodation, and analyses related to accommodation controlled for child anxiety.

Results: Both child anxiety and child-reported parental accommodation moderated the interaction between age and task condition on right amygdala activation to fearful faces (anxiety: $b = -0.01$, $SE = 0.004$, $pFDR = 0.009$; accommodation: $b = -0.01$, $SE = 0.002$, $pFDR = .010$). Within the Alone Condition, there was a negative association between age and amygdala activation in youth with low anxiety symptom severity ($b = -0.05$, $SE = 0.02$, $p = 0.002$) and accommodation ($b = -0.05$, $SE = 0.02$, $p = 0.005$). There was a similar, albeit weaker, association for youth with average anxiety symptom severity ($b = -0.02$, $SE = 0.01$, $p = .033$) and accommodation ($b = -0.03$, $SE = 0.01$, $p = .024$). However, there was no association between age and amygdala activation in youth with high anxiety symptom severity or accommodation ($ps \geq 0.749$). Within the Parent Condition, there was no significant association between age and amygdala activation regardless of anxiety symptom severity or accommodation ($ps \geq .067$). We observed a similar three-way interaction on left amygdala activation, but only for child anxiety symptom severity ($b = -0.01$, $SE = 0.004$, $pFDR = .020$).

Only child-reported accommodation moderated the interaction between age and condition on right vmPFC activation ($b = 0.02$, $SE = 0.01$, $pFDR = .001$). Within the Alone Condition, there was no association between age and vmPFC activation regardless of accommodation ($ps \geq 0.061$). Conversely, in the Parent Condition, age was only negatively associated with vmPFC activation in children who reported low accommodation ($b = -0.07$, $SE = 0.04$, $p = 0.049$); this association was not significant in children who reported average or high accommodation ($ps \geq .074$). We observed a similar three-way interaction on left vmPFC activation

($b = 0.01$, $SE = .01$, $pFDR = 0.010$). There were no significant associations with parent-reported accommodation ($pFDR \geq 0.241$).

Conclusions: Age-related patterns of neural reactivity either alone or in the presence of a parent differ depending on child anxiety and child-reported parental accommodation. While cross-sectional, these results may reflect expected developmental patterns of increasingly independent amygdala regulation and reduced reliance on parents for vmPFC engagement during fearful face processing in children with lower anxiety severity and lower child-reported accommodation. On the other hand, children with greater anxiety symptom severity and greater accommodation do not show these expected developmental patterns. This may reflect a pathway by which anxiety and accommodation interfere with developmental patterns of increasingly independent fear regulation.

Keywords: childhood anxiety, fMRI, anxiety, threat, Parental Factors

Disclosure: Nothing to disclose.

P58. LONG-TERM PARTNERS IN THE THERAPEUTIC JOURNEY FOR ANXIETY-DEPRESSIVE DISORDERS: THE SYNERGISTIC ROLE OF PSYCHOLOGIST AND PSYCHIATRIST

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Background: In a socio-cultural context marked by accelerating change, uncertainty, and constant exposure to stressors, anxiety-depressive disorders (ADD) represent a major global public health challenge. ADD are a heterogeneous group of psychiatric conditions characterized by the coexistence of affective and anxiety symptoms, with a significant impact on daily functioning and quality of life. Global prevalence is estimated at 10–15%, with higher incidence in working-age populations and in individuals exposed to chronic stress. According to the World Health Organization (WHO, 2024), approximately 4% of the global population suffers from anxiety disorders, while 3.5–4.4% are affected by depression, both representing a substantial global health burden. In the absence of integrated care, ADD are associated with increased risks of chronicity, comorbidities, reduced life expectancy, and relapse. International guidelines (NICE, APA, WPA) therefore recommend a psychologist-psychiatrist partnership that combines pharmacological treatment, psychotherapy, and psychosocial interventions. Effective management requires not only punctual collaboration, but a long-term interdisciplinary alliance, integrating psychotherapeutic and pharmacological approaches to support not only symptom remission but also personal meaning reconstruction and strengthening of internal resources.

Methods: We conducted a narrative review of 42 scientific articles published between 2020 and 2024, selected from PubMed and Scopus, including randomized controlled trials, meta-analyses, and international practice guidelines (NICE, APA, WPA). These data were supplemented by an observational study of 80 patients (aged 18–65 years), diagnosed according to DSM-5 and ICD-11, followed for 12 months in both outpatient and inpatient settings.

The integrated intervention targeted four dimensions. The four dimensions were selected based on recent scientific evidence emphasizing the value of multimodal approaches in ADD: (1) biological – personalized pharmacotherapy with adherence monitoring and neurobiological adjustment; (2) cognitive-behavioral – CBT, ACT, cognitive restructuring, graded exposure, emotional regulation; (3) existential-narrative – identity reconstruction, value clarification, coherence; (4) transgenerational – restructuring inherited emotional patterns via experiential psychotherapy, psychodrama, and somatic techniques.

Efficacy was assessed using internationally validated psychometric instruments (HADS, PHQ-9, STAI, MINI International Neuropsychiatric Interview), semi-structured clinical interviews, and reflective journals kept by patients, analyzed at baseline, 3 months, and 12 months. The methodological approach aimed not only at symptom reduction but also at enhancing global functioning and long-term psychological resilience.

Results: Clinical data and qualitative observations revealed a mean reduction of 46% in anxiety and depression scores at 3 months and 61% at 12 months, with a significantly reduced risk of relapse. Compared to pharmacological monotherapy, the integrated approach achieved higher remission rates at 12 months (+18%) and lower relapse rates (−22%). In comparison with psychotherapy alone, symptoms remitted more quickly (1.5 months). Cost-effectiveness analysis (QALY) confirmed the economic advantage through reduced relapses and hospitalizations, supporting the feasibility of implementing this model in wider clinical practice. Patients reported improved self-regulation, resilience, and greater clarity in life roles and goals. The relational dimension of the intervention enabled reinterpretation of traumatic experiences from an integrative perspective, mitigating negative transgenerational impact. Subjective feedback indicated increased autonomy and life meaning, strongly correlated with sustained remission. Three key mechanisms underpinned these outcomes: enhanced sense of personal control, normalization of experiences through empathic validation, and consolidation of support networks. Beyond psychometric improvements, patients demonstrated tangible life changes, such as returning to work, resuming social engagement, and reconnecting with family.

Conclusions: Psychologist–psychiatrist collaboration represents an optimal approach to treating ADD by addressing biological, cognitive, emotional, and relational dimensions simultaneously. The results support this model as effective not only in symptom remission but also in transforming patients' subjective experiences, preventing relapses, and improving quality of life.

Integration of these interventions increases not only clinical success rates but also the sustainability of outcomes, offering a feasible and cost-effective model for healthcare systems. Instead of focusing exclusively on difficulties, patients learn to identify and activate inner resources, turning therapy into a journey of reconstruction and personal growth.

The therapeutic journey of a patient with anxiety–depressive disorder does not follow a linear path and lacks predetermined “instructions.” However, through consistent interdisciplinary collaboration, specialists can accompany the patient in discovering sustainable ways of understanding and managing their own life and in maintaining long-term emotional balance.

Keywords: Anxiety and Depression, Clinical interventions, cognitive

Disclosure: Nothing to disclose.

P59. PERIPHERAL LIPID DYSREGULATION AND VASCULAR DYSFUNCTION LINKING ANXIETY TO ALZHEIMER'S DISEASE RISK

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Background: Anxiety and its phenotypes are risk factors for several major diseases of aging including cardiovascular and autoimmune diseases, as well as Alzheimer's Disease (AD) and related dementias (ADRD). However, little is known about the mechanisms underlying the association between anxiety and ADRD risk. Leveraging lipidomics and proteomics in association

with neuroimaging markers allows for the investigation of neurobiological markers contributing to ADRD risk among older individuals with specific anxiety phenotypes (rumination, global anxiety, and worry; RAW).

Methods: We integrated lipidomic profiling, ADRD blood-based biomarkers, and neuroimaging in a longitudinal cohort of older adults (aged 60 years or older). Anxiety phenotypes - rumination, anxiety and worry (RAW) - were classified as High RAW (Penn State Worry Questionnaire (PSWQ) ≥ 50 and/or Rumination Subscale from the Response Style Questionnaire (RSQ) ≥ 50 and/or 17-item Hamilton Anxiety Rating Scale (HARS) ≥ 14) or Low RAW. Cross-validated Elastic Net regression and traditional regression models were applied to investigate associations between NULISeq CNS Disease Panel 120, untargeted-lipidomics, Simoa ADRD blood-based biomarkers, neuroimaging markers, and clinical outcomes. All models were adjusted for age and sex, APOE4 genotype, and normal appearing white matter's T1/T2 ratio when appropriate. Multiple testing was controlled using Benjamini–Hochberg (FDR < 0.05).

Results: Using regression models, we demonstrated that in the High RAW group ($n = 27$), lower levels of plasma amyloid- β (A β) 42/40 ratio (standardized $\beta = -0.665$, $p < 0.001$, FDR < 0.05) and lysophosphatidylcholine (LPC 20:5; $n = 15$; standardized $\beta = -0.068$, $p < 0.001$, FDR < 0.05) were significant associated with higher tortuosity in the internal carotid artery, consistent with changes observed in preclinical AD. While higher plasma neurofilament light (NfL; standardized $\beta = -0.142$, $p < 0.05$, FDR < 0.1) tracked with vascular changes based on T1/T2 ratio in white matter hyperintensities (WMH) in participants with lower anxiety severity (Low RAW; $n = 57$). In a subset of our cohort, plasma of 36 participants at baseline (15 High RAW and 21 Low RAW) were analyzed for lipidomics and inflammatory and endothelial damage markers. We identified a subset of inflammatory (contactin-2, TAFAS, IL-5, IL-9, CCL11, S100A2) and endothelial damage (SAA1, VCAM1) markers significantly associated with worry and rumination, but not global anxiety. Additionally, lipidomic profiling in plasma revealed multiple differential lipids that were substrates (phosphatidylcholine-PC, phosphatidylethanolamine-PE; $p < 0.05$) and products (LysoPC-LPC, LysoPE-LPE; $p < 0.05$) in the Lands Cycle for acyl-chain remodeling associated with RAW, adjusting for sex and age. Given the smaller sample, we focus on effect sizes and report $\beta > 0.2$.

Conclusions: Converging evidence across plasma lipids, ADRD blood-based biomarkers, and neuroimaging points to an ADRD-relevant signature in anxiety phenotypes—most pronounced in High RAW. Lower plasma A β 42/40 and LPC (20:5) related to greater internal carotid artery tortuosity, while NfL tracked alterations within WMH, implicating early neurovascular change. Lipidomics indicated membrane remodeling (decreased PC/PE; increased LPC/LPE) and, together with inflammatory/endothelial markers, tied specifically to worry/rumination. Together, these results suggest that anxiety phenotypes in older participants engage distinct biological pathways, in which lipid dysregulation and ADRD blood-based biomarkers engages vascular–inflammatory pathways that may heighten vulnerability to ADRD. Further research is needed to clarify these biological pathways and explore targeted prevention strategies.

Keywords: Anxiety, Worry, Alzheimer's disease, Lipidomics, Blood-based biomarkers

Disclosure: Nothing to disclose.

P60. PUBERTAL CAUSES OF ADOLESCENT ANXIETY

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Background: Anxiety disorders are the most prevalent mental health conditions in adolescents, and they are associated with significant and long-term impairment across school, social, and home-based settings. However, we do not fully understand why specific anxiety disorders develop in adolescents. While pubertal onset has shown a correlational association with anxiety development, there are many potential confounding variables that limit causal inference.

However, new methodological innovations offer a solution to this challenge. The recent development of targeted maximum likelihood estimation (TMLE) can achieve causal inference even in the presence of nonrandomized observational data. In this study, we evaluated how pubertal onset after ages 9–10 can cause the onset of anxiety two years later.

Methods: To test our hypotheses, we used data from the Adolescent Brain Cognitive Development Study (ABCD). The ABCD baseline cohort (N = 11,876; ages 9–10; 47.8% female) includes a nationally representative longitudinal sample. Hormonal pubertal measurements included salivary samples of DHEA, testosterone, and estradiol (in females only). Physical pubertal development was evaluated using the Pubertal Developmental Scale (PDS), which has been validated against Tanner development assessments. We operationalized anxiety by using the Child Behavior Checklist (CBCL) Anxiety Disorders DSM-Oriented Scale. Confounding variables included ABCD measures of status and stress related to peers, families, academics, race/ethnicity, and socioeconomic status. To assess for the causal role of changes in the aforementioned pubertal variables on changes in anxiety, we used TMLE (see statistics section for more detail).

Results: DHEA and early pubertal development in girls provided significant average treatment effects (ATEs) at the $p < 0.05$ level. All observed effects were in the small range (Cohen's d values of 0.15–0.25).

Conclusions: We provide evidence that changes in both hormonal and behavioral measures of puberty can lead to the development of anxiety in early adolescence. This work provides mechanistic insight that can inform future treatment targets. It also provides methodological precedent that can be used in the development of other types of psychopathology when randomization is not possible.

Keywords: Anxiety, Puberty, Adolescent

Disclosure: Nothing to disclose.

P61. TREATMENT PATTERNS FOR NEWLY DIAGNOSED GENERALIZED ANXIETY DISORDER (GAD): INSIGHTS FROM REAL-WORLD EVIDENCE

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Background: Generalized anxiety disorder (GAD) is a chronic and debilitating mental health disorder characterized by excessive worry, which often causes significant distress that disrupts daily activities, academic or occupational performance, and personal relationships, placing a substantial burden on patients' lives. Current treatments frequently yield suboptimal response and are associated with undesirable adverse events. A systematic literature review (SLR) was conducted to evaluate the current state of the literature examining treatment patterns in GAD. To address gaps in the current literature, a subsequent claims analysis was

completed to characterize the treatment patterns of newly diagnosed GAD patients.

Methods: An SLR was conducted to identify publications on GAD treatment patterns from January 2004 to December 2024 in PubMed, MEDLINE, and Embase. In parallel, a retrospective analysis was performed utilizing closed claims data from the Komodo Healthcare Map™, a longitudinal, deidentified, US pharmacy and medical claims database, to identify and follow patients with GAD. The claims analysis included adults ≥ 18 years old with continuous enrollment two years before and after their first GAD encounter from 2021 to 2024. Patients with a claim for a GAD diagnosis in the two years prior to their incident GAD diagnosis were not included in the analysis. Pharmacotherapy treatment rates and patterns (persistence, discontinuation, switch, and combination) were analyzed for the 12 months following initial GAD diagnosis or treatment. For the treatment patterns group, patients with any GAD-related therapy claims within the 6 months before the index date were excluded.

Results: Four studies have reported treatment patterns for GAD in the US. These indicate low rates of treatment, modest medication adherence, and disparities in access to GAD care. These studies focus on patterns of psychotherapy and medication, but offer limited insight into factors driving utilization patterns or longitudinal treatment outcomes. Ex-US, 54 studies examined patterns of GAD treatment. Most of these studies indicated that selective serotonin reuptake inhibitors (SSRIs), benzodiazepines, and atypical antidepressants (e.g., bupropion, mirtazapine) were frequently prescribed. Some studies addressed specific aspects of treatment, such as the undertreatment of patients by general practitioners (Olsson et al.) and common treatment barriers in GAD (Basile et al.). Based on our SLR, this research is the most comprehensive assessment of recent, real-world US treatment patterns, and one of the few papers in GAD that evaluate treatment switching and discontinuations.

The claims analysis included 208,002 patients with GAD who initiated pharmacotherapy. Within 12 months of diagnosis, 76% received pharmacotherapy, most commonly with SSRIs (45%), followed by benzodiazepines (22%) and atypical antidepressants (21%). Treatment patterns were further analyzed at a drug-class level for 59,275 patients. Following treatment initiation, 18% of patients persisted with initial treatment, 58% discontinued treatment, and 24% experienced a treatment change (switching or combining additional therapy). The median number of days until the first modification in treatment was 87 for those discontinuing, 54 for those transitioning to combination therapy, and only 35 for those switching treatments. An average treatment gap of 208 days (median: 145 days) was observed in patients who discontinued therapy without an immediate switch. Among the 24% of patients with a treatment change, 76% experienced another change in therapy within the study period.

Conclusions: Despite a general lack of US-specific research on GAD treatment patterns, this review underscores substantial unmet needs in current treatment approaches. High rates of discontinuation, undertreatment, and barriers to treatment reflect the inadequacy of available treatment options. The real-world claims analysis bridges some of the gaps in the extant literature by providing an updated evaluation of the treatment burden, highlighting frequent treatment changes and gaps in treatment after discontinuation. These findings emphasize the need for reevaluation of the current GAD treatment paradigm. Further research and innovation to improve the quality and efficacy of GAD pharmacotherapy treatment options are critical.

Keywords: GAD, generalized anxiety disorder, Anxiety and Depression, anxiolytics, Real World Data
Disclosure: MindMed, Employee, Self.

P62. POPULATION CODES FOR ANXIETY IN THE VENTRAL HIPPOCAMPUS

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Background: The ventral hippocampus is a critical node in the distributed brain network that controls anxiety. Anxiety is an emotional state of anticipation of perceived future threats, that in adaptive conditions can enable the subject to protect itself and avoid harm, enhancing its chances of survival. In humans, excessive, persistent, and disruptive anxiety states are clinically diagnosed as anxiety disorders, which differ from one another in the types of precipitating stimuli or situations, but all interfere with daily functioning and are subjectively distressing. States of anxiety are characterized by cognitive patterns and somatic arousal that promote vigilance and avoidance behaviors, and the later can be observed across animal species. The underlying internal state, or its neural representation, can start to be approximated using animal models and recent tools for live imaging of neural populations. Here we used a mouse model to study neural population codes of anxiety in the ventral hippocampus.

Methods: In this study we used miniature microscopy and calcium imaging, to record ventral CA1 (vCA1) neurons in freely moving mice as they explored variants of classic behavioral assays for anxiety-like behavior. Variants of the open field test, elevated plus maze, and elevated zero maze, were used with different light intensity levels, to measure the degree to which an animal internally balances the drive to explore a novel area and the desire to avoid danger in exposed or brightly illuminated regions of the maze. We utilized unsupervised behavioral segmentation to characterize behavioral motifs in these tasks and their underlying vCA1 neural population code.

Results: Unsupervised behavioral segmentation revealed clusters of behavioral motifs that corresponded to exploratory and vigilance-like states. We discovered multiple vCA1 population codes that represented the anxiogenic features of the environment, such as bright light and openness, as well as the moment-to-moment anxiety state of the animals. These population codes possessed distinct generalization properties. Neural representations of anxiogenic features were different for open field and elevated plus/zero maze tasks, showing a stimulus-specific population code. Conversely, neural representations of moment-to-moment anxiety state were similar across both experimental contexts indicating that mice experience anxiety-related states in all compartments of the test rather than exclusively in the anxiogenic compartments. Furthermore, we characterized how behavioral motifs and neural code were affected by pharmacological manipulation.

Conclusions: Our results suggest that anxiety is not tied to the aversive compartments of these mazes but is rather defined by a moment-to-moment behavioral state and its corresponding population code that generalizes across environments.

Keywords: ventral hippocampus, CA1, calcium imaging, Neural decoding, anxiety state

Disclosure: BGB Group, Employee, Spouse/Partner.

P63. IDENTIFYING THE NEURAL ENSEMBLES MEDIATING FEAR GENERALIZATION ACROSS THE LIFESPAN

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Background: A core symptom observed in anxiety disorders and post-traumatic stress disorder (PTSD) is increased fear generalization, as defined by the overgeneralization of fear from a conditioned, fear-inducing stimulus to neutral stimuli, leading to maladaptive fear responses in a safe environment.

We previously reported that adolescent male, but not adolescent female mice overgeneralize fear, while adult female, but not adult male mice overgeneralize fear. Moreover, in aged mice, both males and females overgeneralize fear. These findings highlight clear age- and sex-dependent differences in fear generalization. However, the neural ensembles mediating fear generalization across the lifespan have yet to be identified.

Methods: ArcCreERT2 x enhanced yellow fluorescent protein (EYFP) male and female mice (5-week-old, 6-month-old, and 24-month-old, $n=12$ per group) were injected with 4-hydroxytamoxifen (4-OHT) in order to tag fear encoding neural ensembles. Five hours after 4-OHT, all mice were administered a 1-shock contextual fear conditioning (CFC) protocol. Five days later, mice underwent the contextual fear discrimination (CFD) protocol to assess fear generalization, receiving 1-shock CFC in context A followed by daily exposure to a similar, neutral context B. After 8 days of CFD, mice were euthanized after exposure to either the aversive context A or the neutral context B. Brain tissue was processed for EYFP and Arc immunolabeling, imaged, and analyzed to identify the neural ensembles mediating fear overgeneralization. Activity patterns were correlated between brain regions to identify how age and sex change fear generalization neural activity patterns.

Results: Five-week-old female, but not male mice discriminate between the aversive context A and the neutral context B by day 6 ($p < 0.01$). Six-month-old male, but not female mice discriminate by day 7 ($p < 0.01$), whereas 24-month-old mice failed to discriminate between contexts even by day 8. These behavioral changes were paralleled by changes in the neural ensembles' reactivation. Specifically, the 5-week-old male, but not female mice and the 6-month-old female, but not male mice show decreased reactivation of the aversive fear ensembles in the hippocampus (HPC) dentate gyrus (DG), CA3, CA1, as well as the piriform cortex (PIR), olfactory tubercle (OT), and ventral posteromedial nucleus of the thalamus (VPM) ($p < 0.05$ for all groups). Similarly, 24-month-old mice also exhibited decreased reactivation of the aversive DG ensembles, suggesting that they might not be able to discriminate between the aversive and the neutral context because of the improper activation of the aversive neural ensemble in the neutral environment, which might result in exaggerated fear.

Conclusions: Our results suggest that sex significantly influences fear generalization behavior, and our data shows that these behavioral differences are linked to alterations in HPC, OT, PIR and VPM fear ensembles in both 5-week-old and 6-month-old mice.

As a future next step, we will build whole-brain networks at different ages with the goal of determining how Age, Sex, and Context impact the fear ensembles mediating fear generalization. These experiments will help identify new nodes in the network

mediating fear generalization so that we can target with pharmacological manipulations to reduce fear overgeneralization.

Keywords: fear generalization, adolescence, aging

Disclosure: Nothing to disclose.

P64. FAST-ACTING ANXIOLYTIC PROPERTIES OF PRUCALOPRIDE, A SEROTONIN TYPE 4 RECEPTOR AGONIST, ARE MEDIATED BY THE VENTRAL HIPPOCAMPUS

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Background: Prucalopride, a clinically approved serotonin type 4 receptor (5-HT₄R) agonist for the treatment of chronic constipation, has recently been shown to produce acute anxiolytic-like effects in mice¹. Since the ventral hippocampus (vHPC) plays a pivotal role in the regulation of anxiety, we investigated its contribution to the anxiolytic effects of prucalopride.

Methods: Male BALB/cJ mice (7–8 weeks, 25–30 g, Janvier Labs, France) received bilateral intra-vHPC infusions of prucalopride (1 µg/side) or diazepam (0.75 µg/side), 30 min before behavioral testing in the Elevated Plus Maze and Novelty-Suppressed Feeding paradigms. To assess receptor selectivity, prucalopride (1 mg/kg i.p. or 0.75 µg/side) was co-administered with GR125487, a selective 5-HT₄R antagonist (1 mg/kg i.p. or 0.75 µg/side). Finally, using AAV.CamKII.GCaMP6s.WPRE.SV40, we performed fiber photometry recordings to examine the impact of prucalopride on the calcium activity of vHPC pyramidal neurons.

Results: In the Elevated Plus Maze, acute intra-vHPC infusion of prucalopride, similar to diazepam, induced rapid anxiolytic-like effects compared to vehicle [one-way ANOVA followed by Fisher's LSD post hoc test, ** $p < 0.01$], as shown by increased time spent and entries into open arms. The ratio of distance traveled in open arms to total distance confirmed that this effect was not attributable to changes in locomotor activity [one-way ANOVA, $p < 0.01$ vs. vehicle]. Pre-treatment with GR125487 blocked the anxiolytic-like effects of intra-vHPC prucalopride, supporting a 5-HT₄R-dependent mechanism. These findings are currently being validated in the Novelty-Suppressed Feeding paradigm. Ongoing fiber photometry analyses are assessing whether prucalopride affects pyramidal neuron calcium dynamics at the onset of its anxiolytic effects.

Conclusions: Our results suggest that acute activation of 5-HT₄Rs in vHPC pyramidal neurons by prucalopride induces a rapid anxiolytic response, highlighting the ventral hippocampus as a critical mediator of its fast-acting properties.

References:

¹Mendez-David I, Vriens R, Denny CA, Hen R, Corruble E, David D. P81. Prucalopridelopride, a 5-HT₄ Receptor Agonist, Induces Rapid Anxiolytic Effects in Mice. ACNP 62nd Annual Meeting: Poster Abstracts P1 – P250, Neuropsychopharmacology volume 48, pages 63–210 (2023).

Keywords: ventral hippocampus, Prucalopride, Potential rapid anxiolytic effects., 5HT₄R agonist

Disclosure: Lundbeck Inc., Advisory Board, Spouse/Partner, University Paris-Saclay and Columbia University, New US Continuation Patent Application No. 17/494,218., Patent, Self, University Paris-Saclay and Columbia University, New US Continuation Patent Application No. 17/494,218., Patent, Spouse/Partner.

P65. STRIATAL DOPAMINE IN THREAT RESPONSES AND ADAPTIVE LEARNING

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Background: Defensive behaviors and adaptive threat learning are essential for survival. The tail of the striatum (TS), the most caudal region of the striatum, integrates sensory and cognitive inputs from cortical, thalamic, and monoaminergic areas. Emerging evidence implicates the TS in processing novel, threatening, and salient stimuli. However, the specific TS neuronal circuits engaged during initial threat exposure and how they adjust with threat learning remain poorly understood.

Methods: We employed fiber photometry with the dLight biosensor to measure dopamine (DA) dynamics in the TS of C57BL/6J mice ($n = 6$ males) in response to a visual looming stimulus (VLS) threat, as well as across repeated exposures over multiple days. For comparison, DA signals were also monitored in the nucleus accumbens (NAc) ($n = 7$ males). To assess postsynaptic dopaminergic signaling, we recorded activity of spiny projection neurons (SPNs) in DA receptor 1 (D1R)-Cre mice ($n = 5$ males, 5 females). Additionally, we used the Targeted Recombination in Active Populations (TRAP2) system to label TS neurons activated during initial VLS exposure and subsequent VLS learning ($n = 3–6$ mice per group, both sexes).

Results: We observed robust DA signaling in the TS during initial VLS exposures, which significantly declined across days (One-way ANOVA; day effect: $F(1.845, 9.227) = 18.38$, $p = 0.0007$). In contrast, VLS did not elicit detectable DA responses in the NAc, suggesting regional specificity in dopaminergic threat processing. Recordings from TS D1-SPNs revealed strong activation during early VLS exposures, followed by a marked decrease with repeated presentations (Two-way ANOVA; day effect: $F(1.367, 10.93) = 7.641$, $p = 0.0133$; sex effect: $F(1, 8) = 0.4711$, $p = 0.5119$). Finally, using the TRAP2 system, we labeled TS neuronal ensembles activated during initial VLS exposure, which were no longer detectable following repeated presentations, indicating their transient engagement during early threat processing.

Conclusions: These findings identify DA signaling and post-synaptic networks within the TS as a critical hub for encoding threat responses and enabling flexible behavioral adaptations to environmental threats. Importantly, this work has broad implications for understanding maladaptive threat processing underlying psychiatric conditions such as anxiety disorders.

Keywords: Dopamine, Visual threat detection, Adaptive Behavior, striatum

Disclosure: Nothing to disclose.

P66. ABERRANT BELIEF-UPDATING IN LATE-LIFE WORRY AND RUMINATION: A COMPUTATIONAL APPROACH FOR DIFFERENTIATING PHENOTYPES OF REPETITIVE NEGATIVE THINKING

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Background: Worry and rumination, two commonly co-occurring forms of repetitive negative thinking (RNT) encountered as core symptoms of late life anxiety and depression, are difficult to treat and

increase the risk of relapse for remitted depressive and anxiety disorders, especially in late life. We propose that dysfunctional belief-updating is a fundamental cognitive mechanism underlying RNT and test this employing a parameterized Bayesian model of belief updating. Under this model, updated beliefs (posterior) reflect a precision-weighted combination of previous beliefs (prior) and evidence from new observations (likelihood). We hypothesized that: (1) RNT would not be associated with overall performance on a belief-updating task compared to the optimal Bayesian model, (2) worry would be associated with greater emphasis on pre-existing information (greater prior weight), and (3) rumination would be associated with a failure to incorporate new information appropriately (low update strength).

Methods: We recruited 116 older participants (age ≥ 50) dimensionally for rumination and worry to perform a belief updating task—the Fisherman task. Worry was assessed with the Penn State Worry Questionnaire and rumination with the Response Style Questionnaire rumination subscale. The Fisherman task consists of 16 blocks where participants must rate how likely it is that a fisherman is fishing in one of two lakes containing differing proportions of black and white fish. The fisherman expresses a preference for the lakes by his position on a dock between the lakes (prior) and then proceeds to catch a series of 5 fish from one of the lakes. Ratings are collected before the first fish is caught (initial prior) and after each fish is caught (posterior). The likelihood is quantified by the proportion of fish in the lake and which color fish is caught. We quantified performance with absolute divergence from the optimal Bayesian model; prior weight was collected explicitly at the beginning of each block; and update strength was calculated by fitting a parameterized Bayes model that quantified the relative weight assigned to prior and likelihood information. Associations with worry and rumination were tested with mixed effects models.

Results: 31 participants were excluded for incomplete task data or inadequate understanding of the task instructions. Participants generally performed in line with the Bayesian model of belief-updating (mean deviation $< 15\%$). Neither worry nor rumination were associated with task performance (worry: $\beta = -0.01$, $p = 0.73$; rumination: $\beta = 0.01$, $p = 0.67$). Contrary to our second hypothesis, worry severity was associated with lower prior weight ($\beta = -0.13$, $p = 0.05$), but rumination was not ($\beta = 0.00$, $p = 0.90$). In line with our third hypothesis, rumination was associated with low update strength ($\beta = -0.23$, $p = 0.04$), but worry was not ($\beta = -0.11$, $p = 0.32$).

Conclusions: In this study, we have shown that our task reliably captures the intended belief-updating behavior. Further, worry and rumination are not associated with performance compared to the optimal Bayesian model, but show unique deficits in belief-updating. Low prior weight for worry may inflate perceived uncertainty, thereby requiring excessive information to adequately update beliefs. On the other hand low update strength for rumination is consistent with a failure to incorporate information, consistent with the common description of feeling “stuck” associated with rumination. These findings form the basis for future work that will more thoroughly characterize aberrant belief-updating in RNT, informing specific therapeutic targets for these symptoms.

Keywords: Worry, rumination, Belief Updating, Bayesian Modeling, mood and anxiety disorders

Disclosure: Nothing to disclose.

P67. EATING A HIGH FAT/HIGH CARBOHYDRATE DIET OR A KETOGENIC DIET, BUT NOT FOOD RESTRICTION, ALTERS SENSITIVITY OF MALE RATS TO MORPHINE

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Background: Recent evidence suggests that in addition to negative metabolic consequences, eating a high fat diet can impact drug sensitivity, especially in the context of recreational stimulant drugs (cocaine and methamphetamine). While high fat/high carbohydrate diets are associated with weight gain, ketogenic diets (high in fat, but very low in carbohydrates) and food restriction have been investigated as approaches for weight loss. We hypothesized that eating a high fat/high carbohydrate diet would increase sensitivity of rats to some of the effects of morphine (e.g., tolerance and withdrawal), but that a ketogenic diet or food restriction would not have the same negative impacts.

Methods: Male Sprague Dawley rats ($N = 47$, $n = 7-8/\text{group}$) were either given free access or restricted access (to maintain rats at 85% of free feeding weight) to a high fat/high carbohydrate chow (60% kcal from fat), a ketogenic chow (90.5% kcal from fat) or low fat chow (17% kcal from fat) for several weeks prior to testing with morphine. Morphine-induced antinociception was assessed using the warm water tail withdrawal procedure. This procedure began with a saline injection, followed by 5 cumulative injections of morphine (0.32–17.8 mg/kg IP) that were administered every 15 min. 13 min after each injection, the latency for rats to remove their tail from hot water baths (40, 50, or 55 °C) was recorded. Additionally, to study the effects of chronic morphine exposure (i.e., tolerance), morphine was administered twice daily for 19 days (3.2–56 mg/kg IP), increasing in $\frac{1}{4}$ log doses every 3 days, followed by an additional warm water tail withdrawal assessment with morphine (3.2–56 mg/kg IP). Next, precipitated withdrawal was evaluated using naltrexone (17.8 mg/kg; SC), and observable signs of withdrawal were evaluated (e.g., weight loss, vocalization, abdominal writhing, lacrimation, chromodacryorrhea, ptosis, teeth chattering, tongue protrusion, salivation, jumping, wet dog shake, rearing, paw biting, paw tremor, and diarrhea). Warm water tail withdrawal latencies were converted to a % of the maximum possible effect and were averaged by each dietary group. Total accumulated withdrawal signs (scored as present/absent) induced by saline or 17.8 mg/kg naltrexone were recorded across 3 observation periods (30, 60, and 90 min). Withdrawal-related weight loss was calculated as the change in body weight from the last day of chronic morphine through the next 5 days, including naltrexone administration. Data were analyzed using mixed model ANOVAs with posthoc comparisons where appropriate.

Results: Morphine-induced antinociception was comparable among rats eating different diets when tested under acute conditions, and after chronic administration all rats developed tolerance to morphine-induced antinociception. Further, rats with free access to high fat/high carbohydrate chow developed greater tolerance than rats eating other diets; however, this was mitigated when rats had restricted access to high fat/high carbohydrate chow. Following naltrexone administration, rats with free or restricted access to the ketogenic chow displayed fewer observable signs of morphine withdrawal and experienced less withdrawal-related weight loss than rats in other groups.

Conclusions: These results suggest that while eating a high fat/high carbohydrate might negatively impact morphine sensitivity by increasing tolerance, this can be mitigated with food restriction. Further, these results suggest that eating a ketogenic diet (even in the absence of overall calorie restriction) might reduce opioid withdrawal severity. These results add to a growing literature demonstrating both positive and negative impacts of diet on individual sensitivity to recreational substances.

Keywords: Morphine sensitivity, high fat diet, Ketogenic diet, opioid withdrawal, opioid tolerance

Disclosure: Nothing to disclose.

P68. DELIBERATIVE BEHAVIORS IN MICE LEARNING A DELAYED MATCH-TO-SAMPLE SPATIAL WORKING MEMORY TASK

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Background: Working memory is the cognitive capacity for temporarily holding information in mind for processing or use, and supports most of our actions and behaviors. In rodents, freely-moving working memory tasks typically follow a nonmatch-to-sample task logic, in which the animal needs to vary its consecutive visits to earn rewards. This logic allows deeply-rooted rodent delayed alternation tendencies to drive behavioral performance. In contrast, delayed match-to-sample task logic opposes innate behavioral tendencies, increasing the utility of deliberation in task acquisition. We hypothesized that the frequency of deliberative behaviors (e.g., pausing, rearing, vicarious trial and error) would correlate with improved acquisition and performance of a novel delayed match-to-sample task.

Methods: Adult mice ($n=9$) were trained in a novel multi-alternative spatial working memory task in an automated, 8-arm radial arm maze with a large, open center. Behavior was recorded and classified using a custom machine learning image segmentation pipeline.

Results: Mice could successfully acquire the novel task, learning to performing well beyond chance. Deliberative behaviors initially increased as mice improved in task accuracy, before decreasing with stable performance. Curiously, after days of stable performance, deliberative behaviors reemerged preceding incorrect choices.

Conclusions: Deliberative behaviors are part of the larger process of behavioral refinement necessary for rodent acquisition of a nonintuitive match-to-sample working memory task. Deliberative behaviors aid in overcoming inherent tendencies during acquisition, before facilitating interrogation of reward contingency stability during stable performance.

Keywords: Behavior, Memory and Learning, Machine Learning Classification

Disclosure: Nothing to disclose.

P69. DISRUPTED SEROTONIN: GLUTAMATE SYNERGY AT THE CROSSROADS OF IMPULSIVITY AND REWARD SENSITIVITY

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Background: Dysfunction in serotonin (5-HT) and glutamate (Glu) signaling within the raphe-accumbens circuit is a driver of the cognitive and/or behavioral dimensions underlying impulse control and reward-related behaviors. Vesicular glutamate transporter 3 (VGLUT3) is localized on heterogeneous neurons which co-release Glu and 5-HT within the raphe-accumbal circuit, presenting a novel target in unmasking disorders and behaviors governed by impulsivity. We hypothesized that an imbalance in nucleus accumbens (NAc) VGLUT3 homeostasis marks high trait impulsivity and concomitant non-homeostatic feeding (i.e. beyond basic caloric needs with hedonic components).

Methods: Outbred male Sprague Dawley rats were identified as high (HI) or low (LI) impulsive using the one-choice serial reaction time (1-CSRT) task. Following behavioral assessments, NAc

synaptosomal VGLUT3 protein levels were analyzed via capillary electrophoresis-based immunoblotting. Accumbal VGLUT3 was knocked down in the NAc and effects on motor impulsivity, feeding behaviors, and body composition analyzed.

Results: HI rats expressed higher NAc synaptosomal VGLUT3 protein vs. LI rats ($p<0.05$); there was a positive correlation between VGLUT3 expression and impulsivity ($r=0.643$, $p=0.02$). Knockdown of NAc VGLUT3 decreased impulsivity and high fat food intake vs. baseline in HI rats ($p<0.05$). There were no differences between % fat mass or % lean mass in control vs. VGLUT3 knockdown in HI rats (n.s.).

Conclusions: These data suggest that in HI rats, higher VGLUT3 may govern neurobiological differences in phenotypic high impulsivity and disordered feeding. Thus, an imbalance in VGLUT3 may shift 5-HT:Glu signaling within the raphe-accumbens circuit and contributes to an aberrant neurobiology underlying disorders characterized by a loss of impulse control.

Keywords: Impulsivity, VGLUT3, Raphe-Accumbens

Disclosure: Nothing to disclose.

P70. CHARACTERIZATION OF "DRY HITTING" AND NICOTINE E-CIGARETTE VAPOR INHALATION IN ADOLESCENT AND ADULT WISTAR RATS

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Background: E-cigarettes are the most commonly used tobacco product among US youths. An additional risk unique to e-cigarettes is "dry hitting", a phenomenon where the e-liquid level in a refillable cartridge gets low enough causing the coil to burn. Dry hit vapor has been shown to produce toxic chemical byproducts including acetaldehyde, ketene, formaldehyde, acrolein, and acetone. Stemming from our ongoing investigation of specialized extracellular matrix proteins, perineuronal nets (PNNs), and the activity-regulated cytoskeleton associated protein (ARC/ARG3.1), we hypothesized that subjects repeatedly exposed to dry hit vapor would exhibit distinct behavioral responses compared to repeated nicotine vapor and differentially alter expression of PNNs and ARC, impacting drug-related conditioning.

Methods: Using a customized system of e-cigarette vapor inhalation (La Jolla Alcohol, Inc.), male adolescent Wistar rats (PND 31–40) were randomly assigned to groups that received nicotine (30 or 60 mg/mL; ~2.5–3 mL/cage), nicotine with dry hits (60 mg/mL; 1.75–2 mL/cage) propylene glycol (PG) vehicle, or air for 30 min daily for 7 consecutive days. A separate group of rats received injections of nicotine (0.4–2 mg/kg, s.c.) or 0.9% saline vehicle. Locomotor activity assessment, thermal antinociception (i.e., tail withdrawal test) were used to confirm acute drug intoxication and tolerance.

Serial collection of whole-brain coronal slices and immunohistochemistry were used to identify Wisteria Floribunda Agglutinin (WFA)-positive perineuronal net structures and ARC-positive brain regions (e.g. amygdala, frontal cortex, and hippocampus).

Results: Nicotine vapor inhalation dose-dependently decreased spontaneous locomotor activity given acutely and produced locomotor sensitization given repeatedly, whereas, "dry hits" significantly potentiated these effects. Repeated inhalation of nicotine vapor inhalation induced antinociceptive tolerance as measured by tail flick latency; however, dry hit vapor did not differ. Nicotine vapor inhalation, with and without dry hits, also significantly increased anxiety-like behavior. Immunohistochemical analyses confirmed that repeated exposure to dry hit vapor significantly decreased WFA+ density in the

central nucleus of the amygdala but not the basolateral amygdala ($p < 0.05$).

Conclusions: Overall, these data confirm that inhalation of dry hit e-cigarette vapor may produce different behavioral responses than inhalation of nicotine vapor alone in rats during adolescence. Dry hitting and regular nicotine in rats may differentially alter extracellular matrix structures and immediate early genes integral to neuroplasticity-related learning and behavior. This may indicate a key mechanism underlying an important vulnerability of naïve or unexperienced e-cigarette usage.

Keywords: Electronic cigarette (e-cigarette), nicotine addiction, Adolescents

Disclosure: Nothing to disclose.

P71. DORSAL RAPHE NUCLEUS ENKEPHALIN MODULATES AVERSIVE PROCESSING

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Background: Chronic pain is a complex disease state associated with comorbidities such as depression, anxiety, and increased risk of suicide. Historically, most analgesic research focused on peripheral and spinal sensory mechanisms despite emotional unpleasantness being a key feature of the human pain experience. The endogenous opioid system can powerfully modulate both analgesia and motivational neural circuits and is therefore well-suited to modulate pain-induced negative affect. Dorsal midbrain nuclei such as periaqueductal grey (PAG) and the adjacent dorsal raphe nucleus (DRN) have been shown to be important sites of opioid action. The PAG is canonically associated with opioid-mediated descending pain inhibition and is desensitized during chronic pain states. The specific role of the DRN during chronic pain has not been characterized despite studies showing opioid activity here also modulating pain and motivated behaviors.

Methods: To investigate the functional significance of DRN opioid signaling we used Cre-dependent CRISPR-Cas9 mediated knockdown in Penk-Cre+ mice or their littermate Cre- controls to disrupt enkephalin production and measured consequent changes in mechanical sensitivity and motivated behaviors ($n = 5-11$ mice/sex) including social interaction and sucrose preference. All behavioral tests were counterbalanced with several days between each test. When possible, mice were tested within subjects. Hplex fluorescent in situ hybridization (FISH) experiments in wild type mice ($n = 3-4$ mice/sex) revealed that enkephalin expression is divided into vGlut2, vGlut3, and vGat subpopulations. We then repeated our CRISPR-Cas9 knockdown of enkephalin in vGAT-Cre+, vGlut2-Cre+, or vGlut3-Cre+ mice to determine whether any of these subpopulations were driving the behavioral effects. Finally, we used a Cre-dependent Gq-coupled DREADD in Penk-Cre+ mice (expected $n = 8$ mice/sex) to test whether modulation of DRN Penk neurons could alleviate spared nerve-injury (SNI)-induced pain and motivational deficits, including progressive ratio breakpoint and sucrose preference. For all studies, male and female mice are being used. All animal procedures were approved by the Washington University IACUC and were conducted in accordance with the National Institutes of Health Guide for the Care and Use of Laboratory Animals.

Results: We found that Cre-dependent CRISPR knockdown of enkephalin in DRN did not affect baseline mechanical sensitivity, but it did produce a hyperalgesic response to low-dose

carrageenan injection compared to controls (Two-Way ANOVA, between subjects, Sidak's multiple comparison; $F(1,29) = 31.09$, $p < 0.0001$; $t(12,19) = 4.783$; Cre- $n = 7M/5F$; Cre+ $n = 11M/8F$). In a social interaction test, Cre- mice investigated a novel mouse more than a novel object (Two-Way ANOVA, within subjects, Sidak's multiple comparison; $F(1,25) = 26.66$, $p < 0.0001$; $t(14,14) = 5.165$; Cre- $n = 8M/6F$) whereas Cre+ mice did not ($p = 0.0743$; $t(13,13) = 2.192$; Cre+ $n = 6M/7F$). Similarly, preference for sucrose was decreased in Cre+ mice compared to controls (Welch's t-test, between subjects, $t(16,9) = 3.670$, $p = 0.0037$; Cre- $n = 11M/5F$; Cre+ $n = 2M/7F$).

Our FISH studies indicate the Penk gene expressing cells compose 20% of the DRN. Half of those cells also express Slc17a6 (vGlut2) and another quarter co-express Slc17a8 (vGlut3), so the majority of these enkephalinergic cells in the DRN are also glutamatergic. The other 25% of Penk-expressing cells co-express the GABAergic cell marker Slc32a1 (vGAT; Data are mean of $n = 3M/3F$ C57Bl6/J mice $n = 1-3$ slices/mouse). CRISPR knockdown of Penk in vGAT-, vGlut2-, or vGlut3- mice was only sufficient to replicate the decreased social interaction (Two-Way ANOVA, within subjects, Sidak's multiple comparison; $F(1,18) = 0.8194$, $p = 0.3773$; vGlut2-Cre+ $n = 5M/1F$; vGlut3-Cre+ $n = 5M/4F$; vGAT-Cre+ $n = 3M/3F$), and this effect did not depend on a particular subpopulation.

Our preliminary results show that after SNI, mechanical allodynia developed in both Penk-Cre+ and Penk-Cre- mice. Administration of the DREADD ligand, deschloroclozapine (DCZ), decreased mechanical sensitivity in Penk-Cre+ mice (Two-Way ANOVA, within subjects, Sidak's multiple comparison; $F(1,7) = 3.879$, $p = 0.0249$; $t(7,7) = 3.333$; Cre+ SNI $n = 3M/4F$; Cre-SNI $n = 1M/1F$). Pilot data indicates that DCZ administration also improves SNI-induced deficits in sucrose preference and motivation in a progressive ratio task. Future work will continue to characterize how DRN enkephalin contributes to motivational and affective components of chronic pain.

Conclusions: These results suggest that DRN enkephalin peptide acts to buffer aversive processing. With the loss of enkephalin from the DRN, aversive responses are enhanced, and normally appetitive preferences are diminished. This effect is specific to enkephalin and does not depend on glutamatergic or GABAergic subpopulations. This suggests that DRN enkephalin may be a potential target for aversive processing disorders, such as chronic pain. Preliminary results suggest that modulation of these neurons improves pain-induced motivational and affective deficits, highlighting the potential for translational chronic pain treatment.

Keywords: enkephalin, Pain, aversion, Dorsal raphe nucleus

Disclosure: Nothing to disclose.

P72. USING PUPIL-LIGHT TO MEASURE PUPIL SIZE IN FREELY MOVING ANIMALS

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Background: Arousal regulates both internal emotional states and outward behaviors. Changes in pupil diameter are commonly used to estimate an animal's level of arousal to study its underlying neural mechanisms. Historically, pupil size measurements have required pointing cameras towards the eye, which is commonly done in head-restrained animals but limits ethologically relevant behavioral testing. Restraining animals

introduces stress that increases physiological signals like pupil size. Performing these measurements in freely moving animals is possible but with bulky head-mounted camera setups. To address these limitations, we have developed a lightweight device named Pupil-Light to accurately track fluctuations of pupil diameter in freely moving mice with minimal impact on naturalistic behavior.

Methods: Pupil-Light is a head-mounted device that measures pupillary changes in awake, ambulatory rodents. Our device works by back-illuminating the eye with infrared light propagating through the skull, and by measuring the amount of diffused light that exits the pupil with a small photodiode, which scales with pupil size. To validate our device, we simultaneously recorded pupil size in freely behaving mice with both Pupil-Light and with a head-mounted camera. We then measured the effect of xylazine and fentanyl on pupil size in freely moving mice. Next, we measured pupil size of mice in the open field test, elevated plus maze, splash test, and tail suspension test to assess how pupil-linked arousal changes across behavioral states and contexts. Finally, we recorded pupil size with Pupil-Light alongside measurement of locus coeruleus activity using fiber photometry.

Results: In our validation testing, the signal from our Pupil-Light device was strongly correlated with a ground-truth recording from a head-mounted camera ($N=4$; Pearson's correlation, $r=0.894$, $p<0.001$). In mice treated with xylazine, we observed a decrease in pupil size versus a pre-treatment baseline ($N=6$, paired t-test, $t(5)=2.996$, $p=0.030$). Conversely, mice treated with low-dose fentanyl (known to elicit locomotor hyperactivity) exhibited sharply increased pupil size relative to baseline ($N=8$, paired t-test, $t(7)=$, $p<0.001$). When testing mice in different behavioral tests, we observed context- and behavior-dependent changes in pupil size. In the open field test, we failed to find a significant difference in pupil size in the center versus corners of the arena ($N=8$, paired t-test, $t(7)=1.237$, $p=0.256$). However, in the elevated plus maze, we saw a significantly higher pupil size in the anxiogenic open arms ($N=5$, paired t-test, $t(4)=5.139$, $p=0.007$). In the splash test, we observed that pupil size was significantly reduced versus a home-cage baseline without sucrose presentation ($N=7$, paired t-test, $t(6)=4.011$, $p=0.007$) and during bouts of grooming behavior following sucrose presentation ($N=7$, paired t-test, $t(6)=6.584$, $p<0.001$). Conversely, we observed an increase in pupil size during the tail suspension test versus a home-cage baseline ($N=6$, paired t-test, $t(5)=4.086$, $p=0.010$), with a significantly higher pupil size during bouts of active struggle versus immobility ($N=6$, paired t-test, $t(5)=2.896$, $p=0.034$). In preliminary testing, we observed that fiber photometry recording of locus coeruleus activity could be performed simultaneously with Pupil-Light ($N=1$ recording).

Conclusions: We developed a new device capable of measuring pupil size in freely moving mice. Our validation experiments show pupillary responses measured by Pupil-Light and a head-mounted camera are closely correlated. Furthermore, changes in pupil size were successfully induced by drug treatment and tracked during changes in behavioral states. Our device is small, lightweight, and compatible with other techniques like fiber photometry, which has been utilized alongside Pupil-Light to assess the link between pupil-linked arousal and locus coeruleus activity. Altogether, our work shows the benefits of Pupil-Light to measure pupillary in mice and paves the way for future applications in humans.

Keywords: arousal, Anxiety and stress, pupillometry, Locus coeruleus

Disclosure: Carolina Instruments, Inc, Founder, Self.

P73. ALTERED MATERNAL CARE AND THREAT RESPONSIVITY IN RATS DUE TO RESOURCE SCARCITY IS RELATED TO CHANGES IN STEROID ACTIVITY

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Background: Postpartum stress is impactful for maternal health and can negatively impact maternal care, impacting offspring. The peripartum period is highly plastic, shaped by hormonal fluctuations and vulnerability to stress. Estrogen receptor α (ER α) in brain regions, including the medial preoptic area (mPOA) and basolateral amygdala (BLA), regulates maternal behavior, yet how environmental stressors alter these systems remains unclear. In this study, we used the limited bedding and nesting (LBN) procedure in rats to mimic resource scarcity during the postpartum period and investigated the changes in maternal care patterns, maternal threat response, and hormonal activity.

Methods: Rat dams and pups were exposed to the LBN procedure from postnatal day (PND) 2 to PND 10, during which they had no enrichment and limited nesting material. In contrast, control dams had access to adequate bedding and nesting materials and enrichment. Prior work indicates that PND 5 is a critical day when dams exhibit the most significant alterations in maternal care. Although altered maternal care has been previously reported, a detailed characterization of the impact of the LBN procedure on maternal behavior and the underlying neurocircuits has yet to be established. We continuously scored the frequency and duration of maternal behaviors, including pup-directed interactions (licking/grooming + nursing pups), nest building, and self-directed (self-grooming + rest spent off-nest + drinking + eating) behaviors twice on PND 5 (1h light cycle and 1h dark cycle). We used the entropy rate, which is a summary measure of the randomness of a distribution and transition probability analysis, the probability of observing a particular behavior following another. To evaluate threat responsiveness, we exposed dams to the light-enhanced startle (LES) task, which measures the startle response to loud, white noise bursts in the absence (PND9) or presence of bright white light (PND10). In rats, the light is an anxiogenic stimulus provoking a bigger startle reaction to the white noise bursts. After LES, brains were processed with immunohistochemistry (IHC) and stained for ER α . To evaluate differences in circulating hormone levels, plasma steroids were quantified with liquid chromatography-tandem mass spectrometry (LC-MS/MS) in a different cohort of dams at PND6. Data were analyzed with Student t-test or the nonparametric Mann-Whitney U test when data failed to meet the assumption of equality of variance.

Results: LBN dams ($n=8$) spent more time than control (CTL, $n=9$) dams displaying pup-directed behaviors (Mann-Whitney $U=10$, $n1=9$, $n2=8$, $p<0.05$, two-tailed, $\eta^2=0.37$) and less time performing self-directed behaviors (Mann-Whitney $U=6$, $n1=9$, $n2=8$, $p<.01$, two-tailed, $\eta^2=0.49$). Specifically, LBN dams exhibited more nursing behavior (Mann-Whitney $U=11$, $n1=9$, $n2=8$, $p<0.05$, two-tailed, $\eta^2=0.34$) and spent more time carrying pups (Mann-Whitney $U=2$, $n1=9$, $n2=8$, $p<0.001$, two-tailed, $\eta^2=0.63$) than control rats. Additionally, the LBN procedure caused the dams to spend less time away from the nest area compared to the control condition (Mann-Whitney $U=6$, $n1=9$, $n2=8$, $p<0.01$, two-tailed, $\eta^2=0.49$). The behavior of mothers in the LBN condition was characterized by a higher entropy rate (1.36 ± 0.09) compared to control dams (1.10 ± 0.077 ;

Mann-Whitney $U = 14$, $n_1 = 9$, $n_2 = 8$, $p < 0.05$, two-tailed, $\eta^2 = 0.26$). LBN dams demonstrated more unpredictable maternal care than control dams as assessed with transition probability analysis. Additionally, LBN dams showed heightened LES reactivity, with earlier startle onset ($t(11) = 4.251$, $p < 0.01$, $\eta^2 = 0.62$) and peak ($t(11) = 2.447$, $p < 0.05$, $\eta^2 = 0.35$) in light in comparison to CRL. ER α cells were reduced selectively in the BLA of LBN dams vs. CRL ($t(11) = 4.467$, $p < 0.01$, $\eta^2 = 0.66$), while most circulating steroid hormones were unchanged except for lower pregnenolone in LBN than CRL ($t(20) = 2.115$, $df = 20$, $p < 0.05$, $\eta^2 = 0.18$).

Conclusions: Our findings suggest that resource scarcity increases maternal care, which may suggest LBN dams are trying to compensate for the limited available resources. However, this care is unpredictable and is evidenced by a higher entropy rate than in LBN than in control dams, an effect that has been previously reported in this model in rats and mice. While it is established that threat circuits are altered in the postpartum period, here we show for the first time that LBN affects this threat processing, increasing LES. The effect, when considered with the increased pup-directed behavior, suggests resource scarcity in the postpartum period increases the processing of relevant environmental cues, indicative of hyperarousal in LBN dams. The BLA is necessary for LES and mediates maternal care. Therefore, reduced ER α expression in this region suggests that estrogen signaling within the BLA modulates the impact of stress on maternal behavior and threat processing. The lack of major differences in peripheral steroid hormone levels at PND6 may indicate that stress does not impact circulating levels of hormones or that only the whole length of LBN (2–9) is required to observe main changes in their levels. Further investigation is needed to confirm these possibilities. These findings highlight mechanisms by which environmental adversity shapes maternal care, with implications for maternal and offspring outcomes.

Keywords: maternal behavior, Sex Hormones, estrogen receptor

Disclosure: Nothing to disclose.

P74. A NEUROPEPTIDE SIGNAL REQUIRED FOR ASSOCIATIVE BEHAVIORS RESPONDS TO INTERNAL STATE AND REGULATES VALENCE PROCESSING

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Background: Learning and memory is often modeled through classical conditioning, which depends on the pairing of a conditioned stimulus-unconditioned stimulus pairing (CS-US). Conditioning can be either appetitive or aversive, depending upon the perceived valence of the US. However, while the presentation of stimuli is discrete, but how they are perceived and processed in under the influence of the animals internal state, such as nutrient status. Thus, disrupted internal state can lead to dysfunctional behavior. Neuropeptides are attractive candidates to couple internal state to valence processing, as they respond to internal physiological states and modulate circuit function extrasynaptically. Here we sought to identify neuropeptides that influenced associative behaviors, and aimed to identify how these peptide signals were themselves regulated by internal state. We set out to identify these signals using *C. elegans*, which have molecularly conserved behaviors and neuropeptide signaling, and a wealth of tools to rapidly manipulate neuropeptide function. In the present study we focused on insulin-like peptides, because of their well-established role in responding to internal state, and conserved regulation of memory across species.

Methods: *C. elegans* maintenance: Wild-type and mutant animals were maintained under standard laboratory conditions and fed the *E. coli* strain OP50 ad libitum. Synchronized populations for behavior assays were generated by standard hypochlorite treatment.

Neuropeptide administration: DNA sequences encoding individual peptides were identified via www.wormbase.org, and were flanked with the endogenous cleavage sites for a peptide processing enzyme. These sequences were codon-optimized and cloned into a vector, and transformed into the feeding *E. coli* strain, OP50. Animals were transferred onto lawns with peptide containing bacteria after development, at least two days prior to behavioral assays.

Behavior Assays: Standard positive olfactory association assays were performed. These assays pair the neutral odorant butanone with food (*E. coli*) so that animals form a positive butanone association. Learning and memory were assayed as a training-dependent increase in preference for butanone as measured by population chemotaxis assays (~100 animals per assay) to obtain a chemotaxis index. Memory performance was calculated by Performance Index (Chemotaxis_Index(trained) - Chemotaxis_Index(naive/untrained)). For all behavioral assays, 10–15 replicates were used and, unpaired student's t-tests with Welch's Corrections, or one- or two-way ANOVA followed by Bonferroni post-hoc tests were performed.

Results: After screening loss-of-function mutants for multiple insulin like peptides, we determined that only one, INS-17, was required for learning and memory ($p < 0.001$). We further confirmed that this neuropeptide was specifically required in the nervous system for learning and memory ability. Using an over-expression by feeding approach, we found that systemic administration of this peptide could improve learning and memory ability in young animals ($p < 0.0001$), and could rescue age-related memory deficits. In investigating why INS-17 is required for learning and memory ability in contrast to closely related peptides, we found that regulation of the INS-17 genomic locus underlies this specificity. We find that INS-17 is extremely responsive to food-deprivation, and is responsible for the memory-promoting effects of brief nutrient deprivation prior to appetitive conditioning. We further found that INS-17 is required for animals to sense that they are nutrient deprived in an aversive conditioning paradigm that pairs starvation with a usually attractive cue.

Conclusions: We found that animals must be able to properly sense their internal state, in this case their nutrient status in order to process the valence of stimuli in associative conditioning tasks. We find that this is regulated by neuropeptide signaling, specifically an individual insulin-like peptide. We propose a model that when internal state sensing is altered by disease, neuromodulation by peptides becomes disrupted, leading to altered associative behaviors. It is likely that individual neuropeptides, even those that are structurally similar, likely respond to distinct cues of internal state and influence specific behaviors.

Keywords: Memory and Learning, Internal state modulation, neuropeptides, *Caenorhabditis elegans*

Disclosure: Nothing to disclose.

P75. DISCOVERING TRANSLATIONAL BEHAVIORAL STRATEGIES OF REWARD PROCESSING IN PROBABILISTIC REVERSAL LEARNING TASKS

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Background: Disrupted reward processing is a key phenotype across numerous psychiatric disorders, underpinning clinical symptoms like anhedonia and impaired motivated behavior, which are not consistently alleviated by current treatments. Preclinical research is important in identifying mechanisms to develop new treatments. Yet, bridging preclinical and clinical settings requires robust translational metrics of reward-related behavior which in turn requires direct comparative studies to uncover similarities and differences in behavioral strategies. Probabilistic reversal learning (PRL) tasks are promising translational tools for examining reward sensitivity, a core dimension of psychiatric disorders, across species, with rodent and human analogues already in use. However, despite task similarity, species-specific cognitive and behavioral strategies pose additional challenges to translation. To address this, we used advanced quantitative methods to uncover hidden structure in behavioral data from PRL tasks in mice and humans to identify the shared and distinct internal states in order to define robust translational metrics of reward processing.

Methods: To identify shared internal states of reward-based decision-making, we employed PRL in humans and mice. Human participants ($n = 26$, 13 females) chose between two abstract symbols with 80% or 20% probabilities of monetary gain or loss; after five correct responses in six trials, each subsequent trial carried a 20% chance of reversal. In the analogous task, mice ($n = 23$, 11 females; 7 weeks old C57BL/6J) completed 13 sessions choosing between two levers delivering chocolate milk rewards with 80% or 20% probability. Lever presses triggered auditory cues and initiated the inter-trial interval, with contingencies reversing after five consecutive choices on the high-probability lever. Behavioral data were analyzed using a generalized linear model–hidden Markov model (GLM–HMM), which incorporates Bernoulli GLM observations, to identify reward-learning states as a function of external covariates. The temporal dynamics of learning strategy in both species were modeled and compared with a linear dynamic model.

Results: In the PRL task, humans predominantly occupied a reward-learning state characterized by strong weighting of the previous reward covariate, with posterior probability rapidly reaching a steady level within a single session. A second, subtler reward state with weaker reward weighting was more prominent early in learning but declined as the reward-learning state took over, reflecting a shift from a naïve to an expert valuation strategy. Alongside these states, humans also expressed a distinct reward-history state, marked by sensitivity to rewards beyond the immediately preceding trial, which remained stable across the session and is consistent with memory for prior outcomes. Interestingly, the reward-related states were highly conserved across species: the correlation between human and mouse GLM weights was 0.99 for the reward-learning state ($p < 0.001$) and 0.87 for the subtle-reward state ($p < 0.001$), highlighting strong cross-species similarity in reward valuation strategies. Mice nevertheless required several sessions to reach the stable occupancy of the reward-learning state achieved by humans in a single session. Dynamic modeling further showed that the logistic growth trajectory of the reward-learning state posterior was well captured in both species (RMSE = 0.03 in humans, 0.05 in mice). However, humans reached steady-state occupancy more rapidly, underscoring a shared strategy achieved on different timescales. In contrast, species-specific history states diverged: while humans maintained a reward-history state across trials, mice instead expressed a choice-history state biased toward repeating past actions, with GLM weights showing a negative correlation across species ($r = -0.6$), reflecting opposing reliance on past rewards versus past choices.

Conclusions: We demonstrate that PRL in mice and humans recruits both reward-learning and subtle-reward states that are conserved across species. Additional species-specific states reflect

species-specific behavioral strategies: humans rely more on cognitive integration of reward history, whereas mice show greater dependence on choice stickiness. By linking these behavioral states to underlying neural activity in mice in ongoing work, we aim to uncover the translationally relevant mechanisms of reward processing that are disrupted in depression and other reward-related disorders, such as addiction. This cross-species framework provides a powerful platform for identifying neural circuit alterations that can inform clinical understanding and treatment development.

Keywords: Translational studies, probabilistic reversal learning, reward learning, Dynamical system model, computational modeling

Disclosure: Nothing to disclose.

P76. NOVEL DOPAMINE D4 RECEPTOR LIGANDS DIFFERENTIALLY SHAPE PSYCHOSTIMULANT-INDUCED RISKY CHOICE PREFERENCE WITHOUT ALTERING BASELINE DECISION MAKING IN RODENTS

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Background: Catecholamine neurotransmitter systems, including dopamine (DA) and norepinephrine (NE), regulate prefrontal cortex (PFC)-mediated executive functions such as complex decision making in situations involving uncertain risk and reward. Perturbations of these systems are implicated in ADHD, substance use disorders, traumatic brain injury, and other neuropsychiatric conditions characterized by maladaptive risky choice behaviors. Among catecholamine receptors that have been characterized for specific roles in modulating risk/reward decision making behavior, the DA D4 receptor (D4R) has been least understood due to lack of suitable pharmacological tools. Novel ligands with higher affinity, selectivity, and mechanistic specificity now enable more precise investigation of D4R function.

Methods: To examine the effects of D4R modulation of baseline risk/reward decision making, FMJ-045 (low-efficacy partial agonist), FMJ-038 (high-efficacy partial agonist), and FMJ-054 (full antagonist), were administered intraperitoneally (1–10 mg/kg) prior to performance of the probabilistic discounting task (PDT), a preclinical assay in which rodents choose between a small/certain reward delivered with 100% probability and a large/risky reward delivered with descending probabilities across consecutive trial blocks. To assess modulation of psychostimulant-induced risky choice, PDT performance was also evaluated following co-administration of each ligand (10 mg/kg) with amphetamine (AMPH; 0.5 mg/kg). Locomotor activity was measured concurrently using the open field test (OFT) to dissociate decision-making effects from general motor activation.

Results: None of the 3 D4R ligands altered baseline PDT performance measures, including choice preference, response latency, magazine latency, omissions, or win-stay/lose-shift behavior, indicating these ligands alone do not affect risk/reward decisions. Consistent with previous reports, AMPH increased risky choice and reduced lose-shift behavior, reflecting reduced sensitivity to prior losses. Both FMJ-045 and FMJ-054 attenuated AMPH-induced risky choice and restored lose-shift behavior, although combination with FMJ-054 also increased response latency, suggesting slowed processing or motor output. The combination with FMJ-038 blocked AMPH-induced risky choice and selectively reduced baseline risky choice and win-stay behavior in high-reward probability blocks, reflecting a disruption of stable value representation that leads to erratic switching from

advantageous choices. AMPH increased locomotor activity, whereas none of the D4R ligands affected locomotion, confirming that decision making effects were independent of motor changes.

Conclusions: Selective D4R ligands modulate psychostimulant-driven increases in risky choice preference without affecting baseline decision making or locomotion, indicating D4R contributions to risk/reward behavior emerge primarily under conditions of elevated PFC catecholamine activity. The full antagonist and low-efficacy partial agonist broadly reverse AMPH-induced risky choice by stabilizing D4R signaling and counteracting overactive mesocortical DA input, with the low-efficacy partial agonist acting functionally as an antagonist. In contrast, the high-efficacy partial agonist may further overactivate D4Rs beyond AMPH-induced levels, disrupting probability-weighted value representation in the PFC and selectively reducing risky choice in high-reward probability blocks. These findings provide mechanistic insight into D4R regulation of probability-sensitive risk/reward decisions and highlight their therapeutic potential for disorders marked by aberrant catecholamine function within the PFC, executive dysfunction, and maladaptive risk-taking behavior.

Keywords: Dopamine D4 receptor, Behavioral Pharmacology, Risky decision-making, Prefrontal cortex, Catecholamine regulation

Disclosure: Nothing to disclose.

P77. VENTRAL PALLIDUM CHOLINERGIC NEURONS SIGNAL REWARD AND TRACK CHANGES IN REWARD VALUE

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Background: Deficits in reward processing are a hallmark of substance use disorders, depression, and schizophrenia. Reward processing relies on a distributed neural network that includes the ventral pallidum (VP), a key basal ganglia node that integrates reward-related signals to guide behavior. Preclinical studies reveal that VP activity is strongly tuned to palatable rewards and reward-predictive cues, dynamically tracking their value. Moreover, VP activity is required for effortful responding, cue-driven reward seeking, and both the consumption and hedonic impact of palatable food rewards. Recent studies have shown that GABAergic VP neurons are essential for positive valence reward-related behavior, while glutamatergic neurons have been linked to avoidance. However, much less is known about the role of cholinergic projection neurons (VP-CPNs), which comprise only ~10% of VP neurons in mice but maintain extensive neuroanatomical connections with critical reward circuits.

Methods: Here, we used fiber photometry to characterize VP-CPN activity in vivo during an instrumental task. To this end, we first delivered an adeno-associated virus (AAV) expressing a calcium sensor (FLEX-jGCaMP7f) followed by unilateral implantation of an optic fiber into the VP of adult male and female ChAT-IRES-Cre mice ($n=18$). Mice were trained on a continuous reinforcement (CRF) schedule, where each of the 60 trials in a session began with extension of a lever, which when pressed yielded a milk reward after a 2-s delay. Recorded VP-CPN Ca²⁺ signals were z-scored and analyzed as time-locked events to the onset of the cue (lever extension), lever press, and reward presentation and retrieval. To determine whether neuronal activity is responsive to changes in reinforcer value, two different tests were performed. First, mice underwent satiety-induced devaluation where mice were allowed to freely consume the milk reward for 30 min prior to CRF testing. These sessions were compared to no-prefeeding sessions. In the second test, we directly altered the

reinforcer properties by diluting the milk reward with water. Mice performed 6 sessions of CRF (100%, 50%, 25%, 10%, 2%, and 0% milk). Both tests were conducted in a separate set of ChAT-IRES-Cre mice expressing a Cre-Off FAS-GCAMP6f AAV ($n=15$) to measure activity in non-cholinergic VP neurons. Behavioral performance and trial photometry data were averaged and expressed as mean $\pm$ standard error of the mean (SEM). Paired t-tests were used to compare 2-group data. One-way repeated measure ANOVA was used to evaluate multiple comparisons.

Results: We found that VP-CPN activity consistently increased above baseline following the lever extension (cue), lever press, and reward in a training-independent manner. The largest phasic increase, however, was observed in response to reward retrievals. Pre-feeding the reward before CRF sessions led to fewer retrievals, longer average press latency and session duration. Unexpectedly, pre-feeding led to significantly larger and more sustained reward-evoked responses compared to no-prefeeding sessions. When we performed the task with reward dilution, we also observed increased press latency and session duration, and fewer retrievals as reward concentration decreased. Unlike in pre-feeding, VP-CPN responses shortly after the reward retrieval were decreased with higher dilutions. Furthermore, non-cholinergic VP neurons showed a small cue response and robust activation in response to lever presses and to reward retrieval. Like VP-CPNs, these cells showed significantly enhanced reward-evoked responses following pre-feeding compared to the no-prefeeding sessions. However, we found no significant difference in reward-evoked responses following changing reward dilutions.

Conclusions: Our results identify VP-CPNs as a distinct neuronal population whose phasic responses to food rewards are robust and flexibly modulated by devaluation through either dilution or satiety. By contrast, non-cholinergic VP neurons were influenced only by satiety. These findings suggest that VP-CPNs dynamically track both reward palatability and state-dependent changes in reward value, positioning them as critical contributors to the reward processing functions of the VP.

Keywords: Ventral Pallidum, cholinergic function, Neural Reward Circuitry, In vivo fiber photometry, reward processing

Disclosure: Nothing to disclose.

P78. LONG-RANGE AMYGDALA-PREFRONTAL ENGRAM PROJECTION SUPPORTS REMOTE FEAR RETRIEVAL

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Background: The ability to generate long-term or remote memories is a critical aspect of adaptive behavior, with deficits in remote memory retrieval being hallmark to several neurological and neurodegenerative disorders, including dementia and Alzheimer's disease. Memory retrieval is supported by select neuronal ensembles active at the time of memory encoding known as engrams, which are located in multiple brain regions. While it is known that engram neurons display preferential structural and synaptic connectivity with other engrams, the behavioral contributions of long-range engram-ensemble projections are less clear. Here we investigated the role of engram neuron ensembles within the medial prefrontal cortex (mPFC) and of basolateral amygdala (BLA) engram projections to the mPFC in supporting remote contextual fear memory retrieval in mice.

Methods: We used viral mapping and activity-dependent tagging combined with transgenics, optogenetic and chemogenetic tools (sample sizes ranging from 7 to 13 animals per group) to tag and manipulate contextual fear activated neurons in the

mPFC, as well as mPFC-projecting fear-activated neurons in the BLA at a remote (28d) timepoint in male and female mice. In the most standard experimental design for these experiments, TRAP2 mice were injected with a Cre-dependent FLPo AAV virus into the mPFC and two weeks later were trained in contextual fear conditioning. Mice were injected with tamoxifen immediately after fear training to trigger FLPo expression in fear activated (engram) mPFC neurons. Two weeks later mice were infused a retrograde AAV with a FLPo-dependent Designer Receptors Exclusively Activated by Designer Drugs (DREADD) construct into the mPFC to trigger expression of the DREADD receptor exclusively in mPFC engrams. 28d after fear acquisition animals were re-exposed to the training context (remote retrieval test) in the presence of the DREADD agonist C21. Additional experiments used variations of the viral tools to target mPFC-expressing BLA neurons. Statistical analysis was performed using Prism 9.1 (GraphPad). Parametric data with multiple comparisons were analyzed using a two-way repeated measures ANOVA, followed by Tukey's or Šídák's Post Hoc test with corrections for multiple comparisons. Comparisons of independent groups were made using unpaired t tests or one-way ANOVA, followed by Tukey's Post Hoc test when appropriate. All data were analyzed for sex differences.

Results: We found that mPFC-projecting engram neurons in the BLA and mPFC are necessary for remote contextual fear memory retrieval. Silencing either ensemble selectively impaired remote fear memory retrieval in adult mice (mPFC: mCherry vs hM4D $p < 0.001$; BLA: mCherry vs hM4D $p < 0.05$), an effect that was absent when silencing an equivalent, but non-fear tagged mPFC or mPFC-projecting BLA engram population. Remote fear retrieval also induced an increase in engram-engram synaptic connectivity between BLA and mPFC. Specifically, AMPA:NMDA ratio was increased in BLA-mPFC engram-engram compared to engram-non engram synapses ($p < 0.05$).

Conclusions: Our findings indicate that remote contextual fear memories are supported by a distributed engram network that relies on BLA-mPFC connectivity. These data suggest that the reactivation of long-range engram projections is a key mechanism underlying the persistence of fear memories, highlighting the importance of leveraging this framework to uncover novel memory encoding and retrieval mechanisms across distributed brain networks.

Keywords: Fear conditioning, TRAP2 mice, engram, Medial Prefrontal Cortex (mPFC)

Disclosure: Nothing to disclose.

P79. DECIPHERING THE HETEROGENEOUS AHN GLUTAMATERGIC NEURONS IN MEDIATING FREEZING AND FLIGHT DEFENSIVE BEHAVIORS

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Background: Animals inhabit natural environment where imminent life-threatening stimuli from predators are common. Adopting defensive responses appropriately to threat proximity is crucial for animal survival. The threat-imminence continuum theory categorizes innate defensive behavior into distinct phases according to the proximity of threat stimuli. In a safe state, mice primarily engage in behaviors such as nest-building or feeding. When exposed to potential predator risks, their behavior shifts to a "pre-encounter" mode, characterized by risk-assessment behaviors including sniffing, cautious locomotion in a stretched posture, and exploratory rearing. As threat imminence escalates and predators

are explicitly detected, animals transition from the "pre-encounter" state to the "post-encounter" phase, rapidly deploying adoptive defensive reactions to evade predators, passive freezing dominates as the primary "post-encounter" defensive response. The highest level of imminence corresponds to the "circa-strike" phase, during which the animal is under direct predatory attack. This phase can be further subdivided into low-proximity (primarily inducing escape and persistent freezing) and high-proximity (primarily inducing defensive attack and tonic immobility). The anterior hypothalamic thalamus nucleus (AHN), a diencephalic brain region, is a part of the medial hypothalamic defensive system. AHN is composed of about 92% expressing vesicular GABA transporter (vGAT) GABAergic neurons, with only about 8% expressing vesicular glutamate transporter 2 (vGluT2) glutamatergic neurons. Previous research has focused on GABAergic neurons in the AHN, leaving the function of glutamatergic neurons long neglected. However, whether AHN glutamatergic neurons play a role in distinguishing threat proximities and execute appropriate defense strategies remain poorly understood.

Methods: To investigate whether AHN glutamatergic neurons are involved in defensive behaviors, we employed a visual looming paradigm—known to induce both passive freezing and active flight defensive behaviors, and performed c-Fos immunostaining. To determine if AHN glutamatergic neurons mediate these defensive behaviors, we used optogenetic activation virus AAV-Ef1a-DIO-ChR2-EYFP to selectively manipulate these neurons. For circuit mapping, anterograde and retrograde virus tracing techniques were applied to investigate the downstream brain regions targeted by AHN glutamatergic neurons. Additionally, fiber photometry recordings were used to monitor real-time neuronal Ca^{2+} dynamics in AHN glutamatergic neurons during the defensive behaviors. Data were analyzed via appropriate t test or ANOVA with Tukey's post-hoc comparisons.

Results: We first demonstrate that AHN glutamatergic neurons are involved in both visual looming-induced passive freezing and active flight responses. Furthermore, we identified two functionally distinct subpopulations of glutamatergic neurons within the AHN, which exhibit different characteristics in spatial distribution, structural connectivity, and function, alongside distinct temporal firing patterns during defense responses. The caudal subpopulation projects to the ventrolateral periaqueductal gray matter (vlPAG), is activated during risk-assessment activity in the "pre-encounter" phase, and mediates passive freezing responses in low proximity contexts of the "circa-strike" phase ($N = 6$, $p < 0.0001$). The rostral subpopulation projects to the dorsal preammyllary nucleus (PMD), is activated upon detection of visual predator stimuli during the "post encounter" phase, and mediates active flight responses in high proximity contexts of the "circa-strike" phase ($N = 8$, $p < 0.001$). Optogenetic activation of PAG-projecting ($N = 6$) or PMD-projecting ($N = 8$) AHN glutamatergic neurons specifically can reverse the stereotypical defensive strategies (flight or freezing) induced by the looming paradigm.

Conclusions: This study elucidates the neural circuit basis by which animals adopt context-appropriate defense strategies in response to varying levels of threat imminence in nature settings. When mice are in an environment with potential survival threat, PAG-projecting AHN glutamatergic neurons are activated during risk-assessment activity, driving freezing behaviors that help reducing detection risk. Conversely, when mice are pursued by predators, PMD-projecting AHN glutamatergic neurons are activated by visual predator threats, prompting active flight defense responses. Our results provide theoretical and experimental foundations for a deeper understanding of the neural basis of animal instinctive behaviors, supporting the conclusion that these two distinct subpopulations precisely modulate innate defense responses through two separate circuitry pathways.

Keywords: Innate defensive behavior, Threat imminence continuum theory, Anterior Hypothalamic Nucleus, Glutamatergic neurons

Disclosure: Nothing to disclose.

P80. ANTIFATIGUE-LIKE EFFECTS OF HYMENAEA COURBARIL EXTRACTS AND ISOLATES

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Background: Natural products are increasingly investigated for their potential to mitigate fatigue and improve physical performance. *Hymenaea courbaril* L., long valued in Brazilian traditional medicine, is consumed as “Selva,” a naturally occurring brownish, wine-like extract (WLE). Although popularly regarded as a tonic for endurance, a remedy for fatigue, and a promoter of sexual vigor, experimental evidence for these effects remains limited. Here, we assessed the antifatigue potential of WLE and its bioactive isolates, procyanidin and caryophyllene oxide, in mice.

Methods: Swiss mice [5–6 weeks, 25–30 g; $n = 10$ (5 males, 5 females)] were randomly distributed into treatment groups prior to receiving oral administration of vehicle (10 mL/kg), WLE (50, 100, and 200 mg/kg), procyanidin (5, 10, and 20 mg/kg), or caryophyllene oxide (5, 10, and 20 mg/kg) once daily for 14 days. On day 14, animals were subjected to forced swim (FS), tail suspension (TS), chimney exhaustion (CE), and rotarod (RR) tests. Following behavioral assessments, post-exercise blood lactate, muscle glycogen, and redox biomarkers (glutathione/glutathione disulfide ratio, catalase, superoxide dismutase, ferric-reducing antioxidant power, 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulfonic acid) scavenging activity, and malondialdehyde) were evaluated. All experimental procedures were approved by the Ethics Committee of the University Evangelica de Goiás (Protocol No. 004/2023) and conducted in compliance with NIH guidelines for the Care and Use of Laboratory Animals. Data were subjected to ANOVA followed by Dunnett's or Bonferroni's post hoc tests and expressed as mean $\pm$ SEM ($p < 0.05$ was considered statistically significant).

Results: WLE and procyanidin, but not caryophyllene oxide, significantly increased active task duration across all performance assays [FS: $F(9,90) = 34.9$; TS: $F(9, 90) = 9.9$; CE: $F(9,90) = 13.8$; RR: $F(9,90) = 23.3$; all $p < 0.05$]. These oral treatments enhanced systemic antioxidant defenses, reduced lipid peroxidation, attenuated lactate accumulation, and preserved muscle glycogen, collectively consistent with improved exercise tolerance.

Conclusions: WLE and procyanidin isolate exert antifatigue-like effects in mice through redox modulation and metabolic preservation. These findings provide mechanistic support for the ethnopharmacological use of *H. courbaril* and highlight its potential as a phytotherapeutic candidate for conditions related to fatigue.

Keywords: *Hymenaea courbaril*, Procyanidin, Antifatigue, Endurance, Mice

Disclosure: Nothing to disclose.

P81. SOCIAL COOPERATION IN RODENTS DRIVES CORTICOLIMBIC PLASTICITY

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Background: Cooperation, a key social behavior requiring continuous appraisal of self and others, presents a promising framework to investigate prosocial-specific neural mechanisms underlying socio-affective disorders. The anterior cingulate cortex (ACC), along with its projections to key limbic areas, such as the basolateral amygdala (BLA) and anterior insula (AI), play a central role in the orchestration of social decision-making through the evaluation of emotional salience, and the modulation of social and affective responses. Here, we developed a novel cooperation task in rodents to study how social coordination modulates cortico-limbic circuits and whether repeated cooperation enhances synaptic plasticity in key nodes within the social brain.

Methods: Male and female Long Evans rats ($n = 10$ –16/group) were trained to press levers for food individually before being paired in a custom-built open-field apparatus that required cooperation for reinforcement. Cooperative learning was assessed through the analysis of 3 metrics: probability of successful cooperation in each session, latency to cooperate following cue onset, and cooperative interpress latency (time between two coordinated presses). PET imaging was performed ($n = 22$) pre- and post-intervention—90 minutes of repeated cooperation or non-cooperative reward seeking. Synaptic vesicle glycoprotein 2A (SV2A) binding potential, a marker of synaptic density, was assessed in the ACC, AI, orbitofrontal cortex, striatum, and amygdala. Behaviors during cooperation, such as time spent in open field of the large arena and social proximity/interactions, were also assessed via multiagent pose tracking ($n = 77$). Following incubation with genetically-encoded calcium indicators GCaMP or jRGECO1, we conducted fiber photometry ($n = 11$ –16) to record real-time activity of ACC neurons projecting to the AI and BLA, respectively, during and after cooperation training, as well as projection-specific designer receptors exclusively activated by designer drugs (DREADDs) manipulation to assess causation ($n = 16$ –24/group).

Results: Following Pavlovian and instrumental conditioning, animals took an average of 4–5 days to learn to cooperate [$F(2.764, 19.34) = 8.455$, $p = 0.0011$]. Female animals took longer to reach cooperative criteria [$t(11) = 3.159$, $p = 0.0091$] based on their average latency to cooperate following cue-onset. Cooperative success was higher with familiar training partners than unfamiliar trained animals [$F(2, 20) = 6.320$, $p = 0.0075$]. Diminished visual access also significantly impaired probability to cooperate [$F(2, 66) = 5.383$, $p = 0.0068$] and cooperative interpress latency [$F(2, 66) = 4.427$, $p = 0.0157$]. Increased social interactions ($r^2 = 0.375$, $p < 0.001$) and social proximity ($r^2 = 0.3377$, $p < 0.001$) led to greater levels of cooperative success, while anxiety-like behavior did not impede cooperative success ($p > 0.05$). Photometry revealed that neural activity in ACC→BLA projections were activated during cooperative pressing, while ACC→AI projections were inhibited. Bidirectional chemogenetic modulation of the ACC→BLA projection vs. ACC→AI projection differentially altered cooperative success [statistics pending histology]. Baseline SV2A binding potential in regions assessed in this study did not correlate with cooperative success [$n = 22$, simple linear regressions, $p > 0.05$]. However, repeated cooperation induced significant SV2A binding potential increases in the insula and amygdala 24-h post-cooperation [$F(4, 73) = 2.660$, $p = 0.0393$], suggesting that a brief behavioral intervention was sufficient to induce durable synaptic changes in the rodent “social” brain.

Conclusions: Here, we introduced a novel semi-naturalistic cooperative task to assess social decision-making behavior where animals received mutual reward only after coordinated lever presses. We found that repeated cooperation in our task enhances social brain plasticity and engages distinct corticolimbic circuits with opposing roles for ACC projections to the amygdala and insula, highlighting its potential as a therapeutic intervention for

socio-affective disorders. These findings provide a compelling mechanistic link between prosocial decision making and neural remodeling, suggesting that structured cooperative interactions could be a powerful, non-invasive therapeutic strategy for ameliorating deficits associated with socio-affective disorders.

Keywords: Social Behavior, Anterior Cingulate Cortex (ACC), Decision Making, basolateral amygdala, Social Competence

Disclosure: Nothing to disclose.

P82. A BRAIN-ENRICHED CIRCULAR RNA BLOOD BIOMARKER CAN PREDICT SSRI ANTIDEPRESSANT RESPONSE

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Background: There is an unmet need for easy to measure biological biomarkers to predict antidepressant response. Currently, one of the major tools, DNA-based pharmacogenomics, is limited by the static nature of the genome. Further, the dynamic and adaptable nature of transcriptomic/proteomic biological signatures are significantly constrained by the fact that the vast majority of such molecules do not cross the blood-brain barrier (BBB).

Circular RNAs (circRNAs) are a subtype of non-coding RNAs generated from non-canonical backsplicing and covalent joining of RNA, including exons and introns of protein-coding genes. They cross the BBB and due to their inherent stability are readily-detectable in blood. CDR1as is a conserved circRNA that is particularly abundant in brain tissues, namely cortical neurons, where it regulates activity-dependent neuronal gene expression and synaptic plasticity.

Methods: We utilized baseline (before treatment) and post-treatment whole blood PAXgene samples from the multi-site NIMH-funded Establishing Moderators and Biosignatures of Antidepressant Response in Clinical Care (EMBARC; NCT01407094; $n = 91$) completed study and the ongoing ERANET NEURON funded Biomarkers of Antidepressant Response (ANTARES; NCT05568823; $n = 35$). Both studies were designed to uncover biomarkers for prediction of response to antidepressants including sertraline (SERT). Response to treatment was defined as improvement of 50% or more in the Hamilton Depression Rating Scale (HAM-D-17; for EMBARC) and the Montgomery-Åsberg Depression Rating Scale (MADRS; for ANTARES) and allowed discrimination of responders (R) from non-responders (NR).

RNA isolation was conducted with the PAXgene PreAnalytiX Blood RNA kit 50 (Qiagen, Hilden, Germany) followed by the Monarch RNA cleanup Kit (New England Biolabs, Ipswich, MA). Reverse transcription of 500ng of total RNA was done with the SuperScript IV First-Strand Synthesis System (Thermo Fisher Scientific). Quantitative real time PCR (qRT-PCR) was evaluated using PowerUp SYBR Green Master Mix (Thermo Fisher Scientific) along with custom designed, validated, and sequence-verified circRNA primers.

To investigate the mechanisms linking CDR1as with antidepressant related pathways we treated mouse cortical cultures ($n = 5-9$ /group) with inhibitors for BDNF, CREB, ERK, PKC (respectively ANA-12 (Tocris); KIX-KD (Sigma); FR180204 (Tocris); GF 109203X (Tocris)) or adult C57BL/6J mice in vivo ($n = 9-12$ /group) with MDL100907 2mg/kg, sulpiride, 25mg/kg, or MK801 0.3mg/kg (all from Sigma). Animal protocols complied with French and European Ethical regulations and were approved by the local Ethical Committee.

Results: In the totality of the baseline samples from the EMBARC cohort (sertraline arm; $N = 91$), we observed a clear distinction between SERT-R and SERT-NR based on baseline CDR1as blood expression ($p = 0.0003$ two-tailed Mann Whitney test). This was specific to this circRNA, since expression levels of circTULP4 and circCRY2, two other brain-enriched circRNAs did not exhibit any difference between SERT-R and SERT-NR.

We validated these findings in 35 whole blood PAXgene samples from the ongoing European ANTARES study. Our results showed a significantly higher expression of CDR1as (2-fold) in baseline whole blood samples in SERT-NR vs SERT-R ($p < 0.001$, two-tailed Mann Whitney test), just as it was observed in the EMBARC cohort. Looking at the pooled data from both clinical cohorts ($N = 126$ patients in total), we observed a significant difference in baseline CDR1as levels between SERT-R and SERT-NR ($p < 0.0001$ with a 1.82-fold higher expression in SERT-NR vs SERT-R).

Focusing on the Placebo arm of the EMBARC study, we measured CDR1as baseline levels in responders (PLA-R) and non-responders (PLA-NR) to 8 weeks of Placebo treatment using the same clinical criteria used for determination of response to SERT. We observed no difference in baseline CDR1as levels in PLA-R and PLA-NR.

We performed a receiver operating characteristic (ROC) curve analysis on the baseline SERT samples and calculated the area under the curve (AUC, an index of potential use as a predictive biomarker), the positive predictive value (PPV), negative predictive value (NPV), and corresponding accuracy of CDR1as as a single analyte. We found an AUC of 0.72 ($p < 0.001$) in the totality of the 91 baseline EMBARC samples examined by using a single cut-off for discriminating between SERT-R and SERT-NR (Ct cutoff that achieved the best sensitivity and specificity in the first EMBARC discovery cohort data; selected and kept locked in all other datasets and analyses). We repeated the AUC analysis in the ANTARES clinical cohort ($N = 35$). Our results showed an AUC = 0.83, $p = 0.0018$ in this independent clinical validation cohort. Collective (EMBARC + ANTARES) data gave the following: PPV = 79.71 (71.5–86; 95%CI); NPV = 75.65 (62.9–85; 95%CI); accuracy = 78.18 (69–85; 95%CI).

In preliminary mouse experiments inhibition of the BDNF, CREB and ERK pathway caused a reduction of CDR1as by 30% ($p < 0.05$), 50% ($p < 0.01$) and 75% ($p < 0.001$) respectively. PKC inhibition had no effect. In vivo inhibition of 5-HT_{2A} receptors resulted in a 40% reduction of CDR1as expression in cortical and subcortical regions.

Conclusions: Blood expression levels of CDR1as circRNA at baseline provided reliable prediction of response to the SSRI sertraline in two independent cohorts. Neuronal/brain CDR1as is regulated by the antidepressant-regulating pathways involving both 5-HT_{2A} and BDNF/TrkB-ERK/CREB.

Keywords: Biomarker Candidates, Antidepressants, circular RNA

Disclosure: Circular Genomics, Advisory Board, Self.

P83. METABOLIC PREDICTORS OF KETAMINE RESPONSE IN TREATMENT-RESISTANT DEPRESSION: THE ROLE OF PLASMA LACTATE

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Background: Ketamine is a rapid-acting antidepressant effective in treatment-resistant depression (TRD), yet variability in clinical response limits its utility. While neurotrophic and inflammatory biomarkers have been explored as predictors of ketamine efficacy, mitochondrial and metabolic markers remain under-investigated. This study evaluates plasma lactate, pyruvate, and their lactate-to-pyruvate ratio as indirect indicators of mitochondrial function, examining whether baseline profiles and acute metabolic shifts following infusion are associated with depressive symptom severity and treatment response.

Methods: Participants were enrolled in the Biomarkers of Ketamine Study, an open-label, single-arm, multi-site clinical trial (NCT03156504) approved by institutional review boards at Johns Hopkins University, Mayo Clinic (Rochester, MN), University of Michigan, and Pine Rest Christian Mental Health Services (IRB #16-009737). Plasma levels of lactate and pyruvate were measured at baseline, 24 h after the first infusion, and 24 h after the third infusion. Depression severity was assessed using the Montgomery-Åsberg Depression Rating Scale (MADRS). Remission was defined as a MADRS score ≤ 9 at 24 h after the third infusion. Response was defined as $\geq 50\%$ reduction in MADRS score from baseline at the same time point. Variables were assessed for normality and transformed or standardized as necessary. Linear mixed-effects models (LMM) were used to analyze repeated-measures data, with random intercepts for individual subjects.

Results: Across all models, patients who showed a decrease in lactate levels over time were associated with a higher probability of treatment response, whereas stable or rising lactate levels were linked to non-response. Plasma lactate levels significantly decreased following infusion 1 (mean change: -0.22 mmol/L, $p < 0.001$) and infusion 3 (mean change: -0.32 mmol/L, $p < 0.001$). Higher lactate concentrations measured 24 h post-infusion were associated with a reduced antidepressant response by the third infusion ($p = 0.002$). Similarly, a higher lactate-to-pyruvate ratio predicted a less robust antidepressant response ($p < 0.001$). In contrast, pyruvate levels were not significantly associated with treatment outcomes. Demographic factors, including sex, age, and BMI, did not significantly influence these associations. Within-session fluctuations in plasma lactate significantly modified the antidepressant trajectory.

Conclusions: A post-ketamine metabolic shift, particularly plasma lactate reduction, emerged as a robust predictor of clinical response. These findings highlight the relevance of mitochondrial metabolism in ketamine's mechanism of action and support the inclusion of metabolic monitoring in personalized treatment strategies for TRD.

Keywords: Treatment-resistant depression, Ketamine, Lactate, Biomarkers

Disclosure: Nothing to disclose.

P84. PLACENTAL INTERLEUKIN-6 PREDICTS INFANT FRONTAL ASYMMETRY, BRAIN GROWTH, AND EXTERNALIZING BEHAVIORS: EVIDENCE FOR SEX-MODERATED RISK PATHWAYS

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Background: Prenatal exposures that alter the intrauterine environment are increasingly recognized as critical determinants of infant neurodevelopmental trajectories. Intrauterine inflammation has been hypothesized to program early risk for psychiatric

disorders and other mental health concerns that emerge later in life. Early indicators of such risk include alterations in neural activity patterns, accelerated brain growth, and disruptions in emotion regulation and behavioral control. Among inflammatory mediators, interleukin-6 (IL-6) is a potent pro-inflammatory cytokine and a promising biomarker of maternal immune activation that has been implicated in altered neurodevelopmental outcomes. While maternal peripheral IL-6 levels during pregnancy have been linked to later child outcomes, the contribution of placental IL-6 to infant brain and behavioral development remains poorly understood. Placental inflammation reflects the local inflammatory milieu within the fetal environment, which may have direct implications for fetal neurodevelopment. IL-6 is one of the only known cytokines to cross the placenta. Accumulating evidence suggests that prenatal risk processes may differ by sex, with male offspring often showing greater vulnerability to intrauterine inflammatory exposures. Establishing placental biomarkers that predict sex-specific developmental risk could inform early identification and prevention strategies for mental health concerns.

Methods: We investigated the associations of placental IL-6 protein levels with infant brain function, brain growth, and behavioral health in a large, prospective cohort ($N = 302$ mother-infant dyads, 49.2% female infants). Placental samples were collected at delivery and assayed for IL-6 protein concentration using standardized immunoassay protocols. Infant brain activity was measured via high-density electroencephalogram (EEG) at one month of age, with frontal asymmetry indices derived from power spectra. Brain growth was assessed using repeated head circumference measurements from birth through 12 months, yielding growth trajectories across the first year. Infant behavioral health was assessed at age two using the Child Behavior Checklist, which provides standardized indices of internalizing and externalizing behaviors. Structural equation modeling was used to test associations between placental IL-6 and developmental outcomes. Analyses included covariates for maternal age, parity, socio-demographic factors, and peripheral IL-6 during pregnancy, as well as infant gestational age, delivery mode, birthweight, and age at assessment. The moderating effects of child sex were explicitly modeled.

Results: For male, but not female, infants, higher placental IL-6 concentrations were associated with increased right frontal lateral asymmetry at one month ($\beta = 0.33$, $p = 0.001$, simple slope $= 0.46$, $p = 0.03$) and greater externalizing behavior problems at two years ($\beta = -0.34$, $p = 0.002$, simple slope $= 2.60$, $p = 0.02$). For all infants, higher placental IL-6 concentrations were associated with greater head circumference growth across the first year ($\beta_s < 0.21$, $p_s < 0.01$). Findings survived correction for covariates.

Conclusions: This study provides novel evidence that placental IL-6 is prospectively associated with early neural activity, brain growth, and externalizing behaviors, particularly in male infants. The findings highlight the placenta as a key biological pathway linking intrauterine inflammation to infant neurobehavioral development. Importantly, the observed sex moderation underscores the necessity of incorporating sex as a biological variable when modeling risk pathways. By identifying placental biomarkers associated with infant brain and behavioral outcomes, this work contributes to a growing effort to improve early detection of developmental vulnerability. Such biomarkers could eventually inform clinical prediction models and guide preventive interventions during critical early developmental windows.

Keywords: Developmental Programming, Placental Inflammation, Behavioral Health, Biomarker, Developmental Psychopathology

Disclosure: Nothing to disclose.

P85. COMPLEMENT AND SYNAPTIC PROTEIN PROFILING IN SCHIZOPHRENIA CSF AND PLASMA: ANALYSIS OF THE PSYCHIATRIC BIOMARKERS NETWORK COHORT

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Background: Schizophrenia (SCZ) is a debilitating condition with limited treatment options that treat the symptoms, not the cause, of the disease. An emerging hypothesis of SCZ cause is excessive synaptic pruning by dysregulated complement protein tagging of synapses. Increased copy number variants of complement C4A gene confers risk for SCZ and increased C4A levels in CSF were found in first episode psychosis patients who developed SCZ. Here we investigated whether there is evidence of excessive complement activation in SCZ and identify biomarkers of complement activity to use for clinical trials.

Methods: We developed a panel of complement, inflammation and neurodegeneration immunoassays, that were implemented on matching plasma and CSF samples in the longitudinal cohort of the Psychiatric Biomarkers Network (SCZ, $n = 33$, healthy controls, $n = 35$; both sexes included). These measures were also analyzed on 2 proteomics panels: SomaScan 7K and Olink Explore HT. We evaluated the biomarker data in the context of cross analyte correlation, CSF:plasma correlation, SCZ vs HC at population level, and outlier analysis.

Results: Elevated levels of plasma C3 were observed in SCZ population ($p = 0.02$), and trends of elevated CSF C3 and NFL were seen ($p = 0.08$ and 0.09 , respectively). A subset of SCZ patients (avg of 20%) showed an increase in C4 and C1s, as well as complement activation markers (iC3b, C1s-C1inh) in CSF. The Somascan panel confirmed much of the elevated complement protein findings and also revealed reduction of synaptic proteins including NPTX2, corroborating previous reports in SCZ. Several of the elevated proteins show good correlation between CSF and plasma ($r = 0.56, 0.42, 0.40$ for C3, NFL and C4, respectively). Levels of most complement activation fragments positively correlated with their parent complement proteins.

Conclusions: The PBN cohort has demonstrated perturbations of several complement, neurodegeneration, and synaptic proteins in SCZ CSF, supporting the hypothesis of complement-mediated excessive neuronal pruning in the SCZ brain. Further biomarker studies are planned on additional samples from our growing longitudinal cohort to further understand SCZ.

Keywords: Schizophrenia (SCZ), biomarkers, complement pathway, Inflammation and cytokines, neurodegeneration

Disclosure: Johnson and Johnson, Employee, Self.

P86. UNIQUE EEG-BASED, ARTIFICIAL INTELLIGENCE/ MACHINE LEARNING (AI/ML) BIOMARKERS FOR PREDICTING TREATMENT RESPONSE IN MAJOR DEPRESSIVE DISORDER AND EPILEPSY

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Background: Currently, the selection of treatments for major depressive disorders (MDD) and epilepsy is hampered by a lack of biomarkers with adequate sensitivity and specificity to predict which patients will derive favorable responses. Delays to identifying effective therapies due to this "trial and error" approach contribute further to disease burden. The electroencephalogram (EEG) offers a practical and promising solution to measure brain electrical activity and presumed underlying neural network circuitry in the clinic setting, and is both non-invasive and inexpensive. We and others have demonstrated that pre-treatment EEG machine learning (ML) biomarkers have the potential to improve treatment selection and optimize outcomes for individual patients with MDD. In order to demonstrate patterns of EEG based predictions in patients with MDD, earlier analyses of sertraline response prediction and of TMS treatment response prediction are presented. Leveraging these same ML methods, we also present an extension of our approach to focal epilepsy (FE), aiming to predict patient-specific response in focal onset seizure (FOS) frequency to anti-seizure medications (ASMs) and to also identify individuals at risk for drug-resistant epilepsy (DRE).

Methods: We conducted a secondary analysis of patients with MDD using EEG data from (1) the EMBARC (Establishing Moderators and Biosignatures of Antidepressant Response in Clinical Care) randomized placebo-controlled trial of sertraline ($N = 215$), and (2) a clinical sample receiving naturalistic (i.e., open-label) TMS treatment ($N = 127$). In both cases, resting-state EEG recordings at baseline (prior to treatment) were utilized. Brain functional connectivity measures, including coherence, power envelope correlation, and covariance, were used as feature sets, and an ML data analytic platform with combined unsupervised (K-means) and supervised learning (XGBoost) was employed to discover response biomarkers for SSRI antidepressant and TMS therapy. Change in HAMD-17 and IDS-SR scores from baseline to treatment endpoint were used as outcome variables for statistical analyses. For focal epilepsy (FE), we analyzed baseline resting-state EEGs and ASM outcomes from 280 patients with new focal onset seizures from 28 hospital systems in the Human Epilepsy Project's (HEP) naturalistic open-label study (3). Information-theoretic and connectivity features (e.g., phase locking value, power-envelope correlation) were extracted. Dimensionality reduction was performed with PCA, data-driven subtypes were defined with k-means, and cluster metrics were incorporated into a Random Forest classifier. Response was defined as seizure freedom for > 12 months or three times the longest pre-treatment seizure-free interval. Model performance was assessed with a nested leave-one-out cross-validation analysis.

Results: For patients who received sertraline in the EMBARC study, an ML-empowered EEG biomarker was identified for prediction of treatment outcome with a balanced accuracy of 81%, 83% sensitivity and 80% specificity, and an AUC of 0.87. For outpatients who received naturalistic TMS treatment in a clinic setting, a distinct biomarker with 79% balanced accuracy was identified, which predicted TMS treatment outcome with 88% sensitivity, 70% specificity, an AUC of 0.89 and with $> 90\%$ consistency. In FOS, distinct models predicted ASM-specific response with strong discrimination. For levetiracetam, the AUC was 0.87, balanced accuracy 75%, sensitivity 90%, and specificity 60%. For lamotrigine, the AUC was 0.81, balanced accuracy 72%, sensitivity 81%, and specificity 64%. A separate model identified (DRE; failure of ≥ 2 adequate ASM trials) with an AUC 0.78, balanced accuracy of 0.68, sensitivity of 0.88 and specificity of 0.47.

Conclusions: Utilizing brain functional connectivity features extracted from baseline resting state EEG data by leveraging AI/ML technology, we were able to predict treatment outcomes after an SSRI antidepressant pharmacotherapy and TMS in patients with

MDD, as have other groups. Extending these methods to FE, our nested models predicted ASM-specific response and identified patients with DRE with high accuracy. The versatility of this ML-powered EEG biomarker approach holds potential for identifying the optimal diagnosis and/or treatment type for individual patients presenting with depression as well as FE, and possibly other CNS disorders. Earlier diagnosis and rapid identification of optimal treatments have the potential to shorten suffering, improve functioning, and lower healthcare and societal costs. Ongoing efforts aim to further test these findings across different treatment modalities for MDD and epilepsy patients. Replication of these data in independent samples, and additional validation methods will also be important in future work.

Keywords: EEG biomarkers, artificial intelligence, Major Depressive Disorder (MDD)

Disclosure: Neumarker, Inc., Consultant, Self.

P87. CHANGES IN EEG NETWORK TOPOLOGY MEASURED BY PERSISTENT HOMOLOGY IN RTMS-TREATED DEPRESSION

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Background: Major depressive disorder (MDD) is a major contributor to global disability, and despite the growing adoption of repetitive transcranial magnetic stimulation (rTMS) as a non-invasive treatment for individuals with treatment-resistant depression (TRD), clinicians lack reliable biomarkers to predict response. Currently, therapeutic benefit is assessed only after 4–6 weeks of daily treatment, delaying clinical decisions and reducing opportunities for early interventions. There is a pressing need for biomarkers that can guide treatment selection and track neurophysiological response early in the rTMS course.

Electroencephalography (EEG) is a widely accessible, cost-effective tool for probing cortical dynamics, but most EEG-based biomarker efforts rely on linear, pairwise connectivity or global spectral metrics, which may overlook the brain's higher-order network structure. In this study, we introduce persistent homology (PH)—a core method from topological data analysis—as a novel method to characterize multiscale features in EEG network organization. Unlike traditional methods, PH tracks how patterns of connectivity emerge, merge, and dissipate across a range of thresholds, revealing clusters (Betti-0), loops (Betti-1), and cavities (Betti-2 and beyond) that are associated with information integration and segregation in large-scale brain networks. These features are inherently multiscale and reflect core aspects of brain organization, including functional segregation and integration. By capturing how these patterns emerge and evolve across a continuum of connectivity strengths, persistent homology enables detection of group-level differences in the brain's capacity for information flow, synchronization, and network reconfiguration—processes that are central to cognitive function and emotional regulation, and highly relevant to depression pathophysiology.

Methods: We analyzed resting-state eyes-closed EEG data from 115 adults (79 females) with MDD undergoing naturalistic rTMS treatment. Data were collected both before and after a complete treatment course (typically 20–30 sessions targeting the left dorsolateral prefrontal cortex). EEG recordings were decomposed into standard frequency bands: theta (4–7 Hz), alpha (8–12 Hz), and beta (13–30 Hz). Functional connectivity matrices were computed using the phase-locking value (PLV) between electrode pairs. PH was then applied to extract topological features, represented as Betti curves—graphs describing the number of

loops or cavities in the network as a function of the PLV threshold (filtration value).

For each subject, we calculated two key metrics from these curves: (1) the peak Betti number, representing the maximum number of topological features of a given type (e.g., loops, cavities), and (2) the filtration value at peak, representing the threshold strength at which the network exhibits peak complexity. Subjects were categorized as responders or non-responders based on PHQ-9 symptom reduction criteria, and group differences in PH metrics were assessed.

Results: Responders and non-responders showed distinct topological patterns. Following rTMS treatment, responders exhibited higher peak Betti numbers in:

- Betti-1, Betti-2, and Betti-3 (theta band).
- Betti-1 (alpha and beta bands).

This indicates increased post-treatment network complexity and integration.

Prior to treatment, individuals who ultimately responded to rTMS showed lower filtration values at peak in:

- Betti-2 (theta band), indicating the formation of enclosed network voids at lower thresholds.
- Betti-1 (alpha and beta bands), reflecting the earlier emergence of two-dimensional loops.

Thus, the networks of responders exhibited topological complexity at weaker connectivity thresholds before treatment.

Conclusions: Our findings highlight persistent homology as a mathematically principled method that captures interpretable network-level predictive and treatment-sensitive features from EEG data. By capturing the “shape” of brain networks across multiple spatial scales and connectivity thresholds, PH provides insights beyond what traditional linear or graph-theoretic measures can offer. Responders to rTMS exhibited early-emerging topological features at baseline and developed more topologically complex network configurations post-treatment—suggesting that both baseline readiness and dynamic capacity for reconfiguration may underlie clinical improvement. The early emergence of topological structure in responders may reflect more accessible or flexible neural architecture, while post-treatment increases in complexity were particularly evident in low-frequency bands associated with attentional and affective processing.

This study is among the first to apply PH to a real-world rTMS cohort and to identify frequency-specific topological biomarkers that may predict outcome. Persistent homology may contribute to earlier treatment stratification and more informed rTMS protocols, pending validation, and deepen the understanding of circuit dynamics in depression.

Keywords: EEG connectivity, Brain Based Markers for Depression, repetitive transcranial magnetic stimulation (rTMS)

Disclosure: Nothing to disclose.

P88. INFLAMMATION AND METABOLIC RISK UNIQUELY PREDICT COGNITIVE IMPAIRMENTS IN ADULTS WITH BIPOLAR I DISORDER: A CROSS-SECTIONAL ANALYSIS FROM THE UCLA BD² SITE

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Background: Cognitive impairment is a core feature of bipolar I disorder (BD-I) and a major determinant of long-term functional outcomes. However, the biological mechanisms contributing to these deficits remain poorly understood. Inflammation and metabolic dysfunction have been implicated in BD-I pathophysiology and may affect cognition through distinct pathways. As part of the BD² Integrated Network Study, this cross-sectional analysis used data from the UCLA site to examine whether systemic inflammation and metabolic risk were associated with cognitive performance and estimated cognitive decline, defined as the discrepancy between current cognition and estimated premorbid functioning.

Methods: Participants were 123 adults (56% female; mean age = 42.2, SD = 13.7) with BD-I, diagnosed using the SCID-5. A total of 103 had complete data for cognitive, inflammatory (high-sensitivity C-reactive protein; hs-CRP), and metabolic (fasting insulin, triglycerides, HDL, LDL, total cholesterol, Apolipoprotein B) variables.

Participants completed a neuropsychological battery comprising 10 standardized T-score tasks: Verbal Fluency (executive functioning); CVLT-III Trials 1–5 (verbal learning); BVM-T-R (visual memory); Trails A (processing speed); Trails B (executive functioning); Stroop Interference (executive functioning); Letter-Number Sequencing (working memory); Wechsler Memory Scale–III (working memory); BACS Symbol Coding (processing speed); and CPT-IP (attention/vigilance), generating standardized scores across six domains and a global composite score. Premorbid cognitive functioning was estimated using the WASI-II Full Scale Composite Score Based on Two Subtests (Vocabulary and Matrix Reasoning), and a proxy for estimated cognitive decline was calculated as the standardized difference between current cognition and estimated premorbid cognition.

Biomarkers were assessed via fasting blood samples. Inflammation was indexed by log-transformed hs-CRP. Two metabolic principal components (metabolic PCs) were derived via PCA: Metabolic PC1 (atherogenic risk: ApoB, LDL, total cholesterol) and Metabolic PC2 (insulin resistance/metabolic dysregulation: insulin, triglycerides, inverse HDL).

Hierarchical regressions tested whether hs-CRP and metabolic PCs improved prediction of (a) global cognition (primary) and (b) a proxy for estimated cognitive decline (secondary), independent of demographics (age, sex, education), mood symptoms (PHQ-9, PMQ-9), and BMI. Exploratory analyses focused on executive function, working memory, and processing speed domains, given prior literature. PCA of the 10 cognitive tasks showed 9 loaded onto a single factor, while Stroop Interference loaded separately, motivating task-level exploratory executive function analyses. Multicollinearity was assessed via variance inflation factors (VIF < 2 for all predictors). Given correlated domains and pre-specified primary hypotheses, no correction for multiple comparisons was applied.

Results: Global Cognition:

hs-CRP improved model fit ($\Delta R^2 = 0.048$, $p = 0.02$), and predicted poorer global cognitive performance in the complete model ($\beta = -0.253$, $p = 0.02$). Metabolic PCs did not improve model fit or relate to global cognition ($ps > 0.15$).

Estimated Cognitive Decline:

hs-CRP improved model prediction ($\Delta R^2 = 0.047$, $p = 0.03$), and was associated with the proxy measure for estimated cognitive decline ($\beta = 0.269$, $p = 0.03$). Metabolic PCs were not related ($ps > 0.45$).

Working Memory:

hs-CRP improved model fit ($\Delta R^2 = 0.041$, $p = 0.03$) and was related to poorer working memory performance ($\beta = -0.247$, $p = 0.03$). Metabolic PCs were not associated ($ps > 0.2$).

Processing Speed:

Metabolic PC2 (insulin resistance/metabolic dysregulation) improved model fit ($\Delta R^2 = 0.034$, $p = 0.03$) and was associated

with slower processing speed ($\beta = -0.280$, $p = 0.03$). hs-CRP and Metabolic PC1 were not related ($ps > 0.35$).

Executive Function – Composite Score:

hs-CRP showed a trend-level effect in improving model fit for executive function ($\Delta R^2 = 0.030$, $p = 0.06$), with a weakly trending association in the final model ($\beta = -0.179$, $p = 0.12$). Metabolic PCs were not related ($ps > 0.15$).

Executive Function – Subtests:

- Verbal Fluency: hs-CRP improved the model ($\Delta R^2 = 0.045$, $p = 0.03$), but did not maintain an independent effect after Metabolic PCs were added ($\beta = -0.148$, $p = 0.20$). Metabolic PC2 strongly predicted verbal fluency ($\Delta R^2 = 0.093$, $p = 0.001$; $\beta = -0.422$, $p = 0.001$).
- Trails B: hs-CRP showed trend-level effects ($\Delta R^2 = 0.030$, $p = 0.07$; $\beta = -0.192$, $p = 0.10$). Metabolic PCs were not related.
- Stroop: neither hsCRP nor metabolic PCs were related.

Conclusions: In this cross-sectional study of adults with bipolar I disorder, higher hs-CRP was independently associated with poorer global cognition and greater discrepancy between current and estimated premorbid functioning, even after accounting for demographics, mood symptoms, BMI, and metabolic risk. Separately, insulin resistance/metabolic dysregulation (metabolic PC2) was associated with slowed processing speed and impaired verbal fluency, indicating an impact on higher-order cognitive efficiency. These findings suggest inflammation and metabolic dysfunction contribute to cognitive impairment through distinct pathways, underscoring the importance of targeting both processes to preserve cognitive function in BD-I and highlighting the need for longitudinal studies to establish causality and inform adjunctive therapeutic strategies.

Keywords: mood disorders, Cognition, Biomarker, inflammation, metabolic function

Disclosure: Nothing to disclose.

P89. METABOLOMIC SIGNATURES OF INTRAVENOUS RACEMIC KETAMINE IN TREATMENT-RESISTANT MAJOR DEPRESSIVE DISORDER: THE BIO-K/NNDC MULTICENTER TRIAL

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Background: While the rapid-acting antidepressant mechanism of action (MOA) of ketamine is not fully characterized, the clinical impact of this first in class novel intervention has been substantial. Pharmacometabolomics provides an opportunity to define pathways underlying ketamine's MOA and investigate variability in treatment response. Prior studies suggest the involvement of mitochondrial energy metabolism and lipid pathways in ketamine response. Building on preliminary work, we used data from the Bio-K trial (NCT03156504), a multisite, open-label study of serial intravenous (IV) racemic ketamine for TRD, to characterize metabolomic signatures of ketamine exposure and explore associations with remission status.

Methods: The Bio-K trial enrolled 75 adults with TRD at four National Network of Depression Centers (NNDC) sites: Johns Hopkins, Mayo Clinic, Pine Rest, and the University of Michigan. This analysis included 69 patients with TR-major depressive disorder. Participants were 18–65 years old, with TRD defined as nonresponse to ≥ 2 prior antidepressant in the current episode.

Exclusion criteria included psychosis, unstable medical conditions, and substance dependence. Participants received three IV ketamine infusions (0.5 mg/kg body weight over 40 or 100 min). Remission was defined as a Montgomery–Asberg Depression Rating Scale (MADRS) score ≤ 9 at 24 h after the third infusion. Metabolomic profiling was conducted using the Biocrates MxP® Q500 kit, quantifying > 600 metabolites across 26 biochemical classes. Quality control steps included exclusion of low-quality samples and metabolites, normalization across plates, log2 transformation, and imputation of values below detection.

All analyses were performed in R version 4.2.2. Mixed-effects models with random intercepts for individuals (lme4 package) evaluated metabolite changes over time (three levels: baseline/pre-infusion 1, post-infusion 1, and post-infusion 3), adjusting for age, sex, and BMI. Differential effects between remitters and non-remitters were assessed via time $\times$ remission status interaction. Log2-transformed metabolite concentrations were modeled as the dependent variable. Estimated marginal means and contrasts (e.g., pre-dose vs. post-infusion) were obtained using the emmeans package (v1.11.1), with p-values adjusted by the Benjamini–Hochberg false discovery rate.

Sensitivity analyses were performed in males and females separately.

Results: Participants were predominantly female ($n = 44$, 63.8%) and White ($n = 66$, 95.6%), with a mean age of 44.1 ± 12.6 years and mean BMI of 28.5 ± 5.5 kg/m². Mean MADRS scores decreased from 27.8 ± 5.8 at baseline to 11.1 ± 9.1 post-third infusion ($p < 0.001$), with 53.6% ($n = 37$) achieving remission.

Early changes (post-infusion 1 vs pre-infusion 1): Ketamine acutely altered acylcarnitines (ACs), with elevations in C0, C2, and C16, alongside reductions in C8, suggesting enhanced β -oxidation and mitochondrial flux. Amino acid (AA) metabolism shifted with decreased aspartate and increased glutamine, glycine, and branched-chain amino acids, consistent with altered neurotransmitter and redox pathways. Biogenic amines (GABA, putrescine) and neuroendocrine markers (cortisol, DHEAS) increased, indicating acute neuroendocrine activation. Metabolic indicators showed elevated GABA synthesis (GABA/glutamate ratio) and serotonin/tryptophan ratios, while nitric oxide (NO)-synthase activity and purine metabolites (xanthine, hypoxanthine) decreased.

Late changes (post-infusion 3 vs pre-infusion 1): Persistent alterations included elevated C16 and C3-DC with reductions in medium-chain ACs (C8, C12, C14), indicating ongoing mitochondrial modulation. Amino acid pathways remained altered, with increased cysteine, glutamine, and histidine, and reduced glutamate and aspartate. Serotonin and putrescine were elevated, while cortisol and DHEAS returned toward baseline, suggesting partial HPA axis normalization. Indicators showed sustained enhancement of GABA, serotonin, and polyamine synthesis, alongside reduced NO-synthase activity and purine metabolism.

Sex-stratified analyses showed similar metabolite changes in men and women. Most metabolite classes showed no significant differences between remitters and non-remitters. However, remitters demonstrated greater increases in GABA synthesis after infusion 3. Mild increases in NO-synthase activity were also observed in remitters.

Conclusions: Ketamine exposure induced broad metabolic changes involving acylcarnitines, AA pathways, neurotransmitter systems, polyamine metabolism, and neuroendocrine markers. Early responses reflected acute mitochondrial stimulation and neuroendocrine activation, while later changes indicated partial adaptation and sustained modulation of neurotransmitter and redox pathways. These findings highlight mitochondrial and neurotransmitter metabolism as potential pathways of ketamine's MOA and underscore the need for further validation of peripheral biomarkers to guide TRD treatment.

Keywords: Depression, IV ketamine, Biomarker, Metabolomics signature

Disclosure: Mayo Clinic, Employee, Self, BD2, Grant, Self, Elsevier, Honoraria, Self, NIH, Grant, Self.

P90. NEUROIMAGING AND AUTONOMIC EFFECTS OF A SINGLE MASSAGE SESSION

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Background: We have previously demonstrated that a structured Swedish Massage (SMT) intervention versus a touch control intervention (LT) has profound acute and longer-term effects on immune function, HPA function, oxytocin and vasopressin in healthy control subjects. We have also demonstrated that 12 sessions of SMT (vs. LT) causes a clinically and statistically significant decrease in both Hamilton Anxiety and Hamilton Depression scores in subjects with generalized anxiety disorder (GAD). For a subset of these subjects the effect was sustained for months to years after the intervention. We have also demonstrated that SMT vs. LT has a profound effect in decreasing fatigue and increasing quality of life in breast cancer survivors with chronic cancer-related fatigue. At this time there are no brain imaging studies investigating the impact of a single (pre-to-post) whole-body massage on brain networks and autonomic balance.

Methods: This study investigated the acute effects of a single SMT vs. LT session on brain circuitry, heart rate and heart rate variability in healthy subjects. We hypothesized that SMT activates two distinct but overlapping neural networks: vagus nerve stimulation of ascending afferent fibers to the brain via the nucleus tractus solitarius (NTS) and C-tactile fibers which enervate hairy areas of the skin. We therefore investigated functional connectivity in the insula, a brain region implicated in these pathways. This study was a 2 arm, randomized, masked study investigating the effects of SMT vs. LT control on brain and autonomic activity in non-psychiatric control subjects. Medically healthy male and female subjects $\geq 18 < 65$ years of age, underwent fMRI followed by one SMT ($n = 15$) or LT ($n = 15$), followed immediately by a second fMRI. Change in resting state fMRI functional connectivity was evaluated in the insula, a brain area implicated in emotion regulation and vagus-related brain circuitry. We also evaluated heart rate variability and pulse during resting state fMRI. Parametric statistics (T-tests, ANOVA) were employed.

Results: Preliminary analyses were conducted in LT ($n = 13$) and SMT ($n = 13$). Resting-state analyses evaluated the acute pre- to post-intervention changes in functional connectivity of insula (uncorrected $p = 0.01$; cluster threshold greater than or equal to 20) associated with SMT relative to the LT control condition. Specifically, SMT produced greater increases in insula connectivity with the precuneus, posterior cingulate gyrus, and temporal gyrus, caudate, frontal pole, and medial frontal cortex, whereas SMT showed reduced connectivity relative to LT with the cerebellum, and occipital fusiform gyrus. Further, Both LT and SMT significantly decreased heart rate ($p < 0.05$) during resting state, however, only SMT increased heart rate variability ($p < 0.05$).

Conclusions: These results are the first indication that a single session of a manualized touch intervention (SMT) vs a touch control (LT) differentially modulate resting-state functional connectivity and autonomic physiology.

These neural changes suggest that SMT preferentially enhances connectivity within circuits supporting interoceptive awareness, self-reflection, and affective integration, and executive control while dampening connectivity in networks tied to cerebellar and occipital regions. This pattern is consistent with the proposed

mechanism of SMT, which engages vagal afferents and C-tactile mechanoreceptors to shift autonomic balance toward parasympathetic dominance. Supporting this interpretation, complementary measures of autonomic function demonstrated HRV (a marker of parasympathetic activity) and decreased heart rate during resting state following SMT. The combination of increased HRV and strengthened connectivity within affective and self-referential circuits points to a mechanism by which SMT may attenuate hyperarousal and improve emotional regulation, processes that are often dysregulated in anxiety disorders. Similarly, reduced reliance on motor and cerebellar networks, together with enhanced relaxation-related connectivity, may underlie SMT's previously demonstrated efficacy in reducing fatigue and improving quality of life. Thus, these acute brain and autonomic effects provide a mechanistic framework supporting the therapeutic use of SMT for conditions characterized by heightened stress reactivity, anxiety, and persistent fatigue.

Keywords: Insula, Heart rate variability, vagus nerve, massage

Disclosure: Nothing to disclose.

P91. SINGLE CELL BLOOD PROFILING REVEALS CELL-SUBSET-SPECIFIC GLUCOCORTICOID DYSREGULATION AND ADAPTIVE IMMUNE ACTIVATION IN DEPRESSION

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Background: Immune dysregulation and glucocorticoid resistance have long been implicated in major depressive disorder (MDD), yet translation into clinical biomarkers has been hindered by the complexity of immune signalling and limitations of bulk transcriptomic approaches. The dexamethasone suppression test, once hailed as psychiatry's first biomarker, demonstrated early promise but was ultimately abandoned due to methodological inconsistencies and the complexity of the test. Genetic studies increasingly implicate adaptive immune cells, particularly activated T cells, in psychiatric pathogenesis across diagnoses (Lynall et al. PMID: 3624372). However, outside of bulk blood studies, there has been little exploration of the peripheral immune transcriptome and cell-subset-specific glucocorticoid dysregulation in depression.

Methods: mononuclear cells (PBMCs) from individuals with current depression ($n = 40$) and matched healthy controls ($n = 37$) recruited in 2020–2023; both sexes were included. Single-cell RNA-seq (3' 10X Chromium) profiled major immune subsets. Analyses comprised unsupervised clustering of transcriptomes, pseudobulk differential expression, and pathway enrichment. To test glucocorticoid dysregulation, depression-associated signatures were compared with published glucocorticoid-response signatures from primary immune cells in a cell-subset-matched manner. We contrasted single-cell findings with our bulk RNAseq meta-analyses of the depression transcriptome and with an in-vitro glucocorticoid resistance assay (dexamethasone suppression of LPS-induced IL-6).

Results: Single-cell analysis revealed significant upregulation of T cell activation signatures in depression ($FDR < 0.05$). Using gene set enrichment analysis, depression-associated transcripts in monocytes, T cells and B cells showed negative enrichment (i.e., were anti-aligned) relative to glucocorticoid-stimulated gene sets, maximal when cell types were matched ($FDR < 0.0001$). These cell-subset-specific effects were not detectable in our bulk blood meta-analysis and were not captured by the ex vivo glucocorticoid-sensitivity assay. We demonstrate convergence

between pathways implicated by genetic risk for depression and transcriptional changes observed in adaptive immune cells.

Conclusions: Our study provides the largest single-cell immune dataset in depression to date, revealing adaptive immune dysfunction and cell-subset-specific signatures of glucocorticoid resistance that are obscured in bulk analyses. The resulting blood-based, cell-resolved signatures provide mechanistic insight and advance the search for clinically actionable biomarkers for patient stratification in depression.

Keywords: Adaptive immunity, Depression, immunopsychiatry, Blood-based biomarkers

Disclosure: Nothing to disclose.

P92. ADVANCED ANALYTICS FOR EEG/ERP BIOMARKERS OF SCHIZOPHRENIA: COMPLEXITY/ ENTROPY, FUNCTIONAL CONNECTIVITY, AND SPECTRAL DYNAMICS

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Background: Resting-state electroencephalography (EEG) and event-related potentials (ERPs) are increasingly recognized as sensitive, translatable biomarkers of brain function. In a previously published study (Cecchi et al. Schizophr Res. 2023 Apr;254:178–189), the EEG/ERP Biomarker Qualification Consortium (EBQC) showed that a variety of resting-state EEG and ERP biomarkers can be measured reliably in healthy volunteers (HV) and patients with schizophrenia (SZ) using standardized equipment and methods. Here, we extend the previous analysis by using advanced analytic methods to assess the effect size and test-retest reliability for measures of complexity/entropy, functional connectivity, and spectral dynamics.

Methods: We use an EEG/ERP dataset from a previously published, industry-sponsored observational study in 161 participants (81 HV and 80 SZ). ERP/EEG testing sessions were performed at Baseline and Retest visits and included four tests: (1) eyes-closed resting-state EEG; (2) auditory active oddball; (3) auditory passive oddball/mismatch negativity; and (4) 40 Hz auditory steady-state response. We analyze EEG/ERP data using several advanced analytic methods, including measures of nonlinear dynamics (SE: sample entropy; MSE: multiscale entropy; LZC: Lempel–Ziv complexity), functional connectivity (PCC: Pearson correlation coefficient; MI: mutual information), and spectral dynamics (ERSP: event-related spectral perturbation). Group differences are assessed using repeated measures ANOVA with group and visit as factors. Effect size is calculated as Cohen's d . Test-retest reliability is calculated separately for HV and SZ groups as intraclass correlation coefficient (ICC).

Results: We report HV and SZ group statistics, group differences, Cohen's d , and ICC across multiple advanced analytic measures (SE, MSE, LZC, PCC, MI, ERSP) and compare them to previously published results based on a conventional analysis of the same dataset (i.e., standard frequency analysis of resting-state EEG and amplitudes/latencies of specific ERP components). Prior quantitative EEG and ERP measures had ICCs typically ranging from good to excellent (0.5–0.8) for both HV and SZ populations. Cohen's d (measuring the difference between SZ and HV) typically ranged from small to moderately-large (0.2–0.6). Entropy measurements detect differences between HV and SZ subjects even when EEG power spectra are similar, i.e., reduced entropy in SZ may reflect impaired network flexibility, relevant in cognitive and negative symptoms. ERSP captures event-locked spectral changes and provides time-frequency resolution capturing transient

dynamics missed in conventional analysis relevant for evaluating sensorimotor integration, deficits in attention and cognitive control.

Conclusions: The present work extends our previous study on EEG/ERP biomarkers of schizophrenia by applying advanced analytic methods. Measures of complexity/entropy, functional connectivity, and spectral dynamics capture nonlinear, multivariate, and multidimensional aspects of EEG/ERP signals. They also provide insight into time stimulus relationships. These provide complementary insights to those from conventional analysis. Combining advanced analytics with conventional analysis of EEG/ERP data has the potential to provide a richer, more comprehensive view of brain dysfunction in CNS disorders and improve our ability to detect the effect of interventions. We discuss the implications of our findings for CNS clinical trials.

Keywords: EEG biomarkers, Disrupted-in-Schizophrenia 1, healthy volunteers

Disclosure: Nothing to disclose.

P93. FRONTOCINGULATE GAMMA POWER AND CONNECTIVITY AS BIOMARKERS OF KETAMINE RESPONSE IN TREATMENT-RESISTANT DEPRESSION

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Background: Treatment-resistant depression affects nearly one-third of patients with major depressive disorder and is associated with prolonged illness duration, diminished quality of life, and elevated suicide risk. In this population, ketamine—a glutamatergic modulator—produces rapid antidepressant effects within hours. Yet clinical response remains variable, with only a subset of patients achieving sustained improvement. This heterogeneity has motivated the search for biomarkers that can identify likely responders in advance and clarify ketamine's mechanisms of action. Because ketamine engages glutamatergic and interneuron-dependent pathways, gamma-band activity has emerged as a candidate biomarker. Gamma oscillatory power reflects cortical excitation–inhibition balance and has been shown to increase following ketamine infusion. Nugent et al. (2019) suggested that the association between gamma power and clinical response may be non-linear, moderated by baseline levels. While these gamma effects were observed across widespread cortical regions, converging neuroimaging evidence highlights the subgenual anterior cingulate cortex (sgACC) and dorsolateral prefrontal cortex (DLPFC) as hubs consistently implicated in depression pathophysiology and as key targets for neuromodulation treatments. Functional and metabolic abnormalities within this frontocingulate circuit have also been linked to ketamine efficacy, suggesting that circuit-level measures may help stratify treatment response. Yet whether baseline sgACC gamma and directional DLPFC–sgACC connectivity jointly determine the likelihood of clinical response remains unknown.

Methods: We analyzed data from 29 adults with treatment-resistant depression who participated in a double-blind, placebo-controlled, crossover trial of intravenous ketamine (0.5 mg/kg). Resting-state magnetoencephalography (MEG) was acquired at baseline and 6–9 h post-infusion. Depression severity was assessed with the Montgomery–Åsberg Depression Rating Scale (MADRS) at baseline and 1-day post-infusion. To quantify treatment response, we calculated a response contrast score defined as the pre–post change under ketamine minus the pre–post change under placebo. Source-localized gamma power (30–50 Hz, log-transformed) was extracted from the sgACC and DLPFC. Effective

connectivity between the two regions was estimated using dynamic causal modeling for cross-spectral densities (canonical microcircuit). Receiver operating characteristics (ROC) analyses assessed the predictive performance of baseline sgACC gamma for the response contrast, and parametric empirical Bayes was used to evaluate connectivity differences and associations with symptom change.

Results: The median antidepressant response contrast between ketamine and placebo was 5 MADRS points. In a multivariable logistic regression model including baseline sgACC and DLPFC gamma, age, sex, and session order, classification performance (area under the ROC curve, AUC) remained significantly above chance across thresholds and reached a stable maximum for contrasts of 7 MADRS points or higher. We therefore defined robust response as a contrast ≥ 7 . Under this criterion, inspection of density distributions revealed that responders clustered at both low and high sgACC gamma values, whereas non-responders were concentrated at intermediate levels. To formally test this pattern, we refit the model with a quadratic sgACC term. Both the linear and quadratic sgACC effects were significant ($p < 0.05$), confirming a U-shaped association, while DLPFC gamma showed only a trend-level effect ($p = 0.07$). The nonlinear model achieved strong discriminative accuracy (AUC = 0.89, 95% CI [0.78–1.00]). In addition, dynamic causal modeling revealed that backward connectivity from DLPFC to sgACC was positively correlated with MADRS response contrast and significantly higher in responders than non-responders by permutation testing ($p = 0.012$).

Conclusions: Baseline sgACC gamma and backward DLPFC–sgACC connectivity independently predicted the antidepressant response contrast between ketamine and placebo. The nonlinear, U-shaped association of sgACC gamma with robust response and the greater top-down connectivity in responders together suggest that both local oscillatory state and circuit-level dynamics shape ketamine efficacy. These findings identify frontocingulate gamma and connectivity as candidate biomarkers for stratifying treatment-resistant depression and provide mechanistic targets for future validation.

Keywords: Treatment Resistant Depression, IV ketamine, magnetoencephalography, gamma power, frontocingulate circuit

Disclosure: Nothing to disclose.

P94. Poster Withdrawn

P95. EFFECTS OF PPAR γ AND GLP1 RECEPTOR AGONISTS ON BRAIN METABOLISM: INSIGHTS FROM NEURON-DERIVED EXTRACELLULAR VESICLES, A PRELIMINARY REPORT

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Background: Insulin resistance (IR) is implicated in central nervous system (CNS) disorders, including depression and Alzheimer's disease (AD). We analyzed biological samples from two cohorts of clinical trial participants: (1) participants who received active pioglitazone compared to placebo (PPAR γ agonist, N = 12) in individuals with unremitted depression after six months of treatment as usual and (2) participants who received liraglutide (glucagon-like peptide 1 [GLP1] receptor agonist, N = 15) compared to placebo in middle-aged people at genetic risk for AD.

Methods: These cohorts were used to assess the effects of pioglitazone and liraglutide on CNS insulin signaling using neuron-derived extracellular vesicles (NDEVs) as biomarkers. These

cohorts were both placebo-controlled cohorts, with previous findings related to peripheral measures of IR. In this study, we utilized available biological samples to measure biomarkers of insulin resistance in NDEVs. Eleven Akt-mTOR pathway proteins were measured before and after 12 weeks of treatment in both groups.

Results: Participants who received pioglitazone experienced broader changes, with significant increases in GSK3 β (Ser9), mTOR (Ser2448), and RPS6 (Ser235/Ser236; all $p \leq 0.02$) compared to placebo, and 77% of participants showed mTOR (Ser2448) response. Participants who received liraglutide demonstrated significantly increased NDEV-associated phosphorylated Akt (Ser473) and mTOR (Ser2448; $p = 0.04$ and $p = 0.025$, respectively) compared to placebo, with 40% and 30% of participants in the liraglutide group showing biomarker response in both Akt (Ser473) and mTOR (Ser2448), respectively. These effects were largely independent of changes in fasting insulin and 120-minute glucose during the oral glucose tolerance test.

Conclusions: Our findings demonstrate distinct, CNS-specific biomarker responses to both PPAR γ agonists and GLP1 receptor agonists.

Keywords: neuron-derived extracellular vesicles, Depression, GLP-1 receptor agonist, PPAR γ agonist

Disclosure: Nothing to disclose.

P96. CLINICAL AND NEUROIMAGING CORRELATES OF NATURAL LANGUAGE PROCESSING-DERIVED SPEECH FEATURES IN CLINICAL HIGH RISK YOUTH

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Background: Extensive research has shown that individuals with psychosis exhibit characteristic speech disturbances, such as poverty of speech, disorganization, perseveration, and illogicality, that often appear in attenuated form during the prodromal period. Detecting these subtle markers may provide indicators of psychosis risk. Prior work using Natural Language Process (NLP) techniques have observed increased pronoun use (especially first-person), reduced clausal complexity, disruptions in noun retrieval (Henry and Crawford, 2005; Ehlen et al., 2023), and decreased use of adjectives and adverbs (Bilgrami et al., 2025; Mota et al., 2014, 2017; Minor et al., 2015). Findings from cognitive neuroscience further link such parts of speech to specific cortical regions—for example, nouns to the left middle and inferior temporal gyri (Indefrey and Levelt, 2004), adjectives to the MTG/ITG/temporal pole (Caramazza and Hillis, 1991), and adverbs to the left inferior frontal gyrus (Friederici et al., 2003). Here, we present the first study to jointly examine language-feature frequencies and parts-of-speech patterns in individuals at clinical high risk for psychosis (CHR) and to compare these measures with cortical thickness in the same participants. We test the hypothesis that reductions in specific language features correspond to disruptions in their associated cortical regions, thereby shedding light on potential mechanisms of psychosis risk.

Methods: We used NLP to identify linguistic features in 611 CHR and 145 healthy control subjects from the Accelerating Medicines Partnership Schizophrenia (AMP SCZ) dataset. The mean subject age at assessment was 21.30 years ($SD = 4.1$). Both male and female subjects were included in this study (59.4% female). Demographic, clinical, cognitive, and structural neuroimaging were collected at baseline, and subjects were followed longitudinally for repeated assessments – including monitoring for conversion to full psychosis. Unstructured interview recordings were transcribed to text using OpenAI's Whisper-

large-v3 model, and pyannote. Speaker role assignment was accomplished using the large language model Llama 3.3, 70B. The software package, Stanza, was used for linguistic feature parsing of language features, e.g. parts of speech, verb conjugations, clause types. In total, 102 linguistic features were extracted for each subject. These language features were subsequently compared with clinical and cognitive measures, as well as structural neuroimaging FreeSurfer outputs (normalized gray matter volume and cortical thickness measures), using regression analyses to identify associations of interest.

Results: As predicted, reductions in the use of specific parts of speech were associated with alterations in gray matter volume. CHR individuals produced fewer nouns (PFDR, $t = 2.8 \times 10^{-2}$), which was linked to thinning of the left inferior temporal pole ($R = -0.13$, $P = 1.1 \times 10^{-2}$). They also showed reduced use of adverbs (PFDR, $t = 6.0 \times 10^{-3}$), which was associated with decreased thickness in the left middle temporal ($R = 0.10$, $P = 4.4 \times 10^{-2}$). In contrast, pronoun use—particularly possessive pronouns—was elevated (PFDR, $t = 1.1 \times 10^{-6}$), potentially reflecting compensation for reduced noun use. These findings parallel prior reports of language disruption in schizophrenia and align with neurobiological evidence linking specific linguistic features to cortical regions. The clinical relevance of these patterns was supported by leave-one-out cross-validation using CHR class probability scores from a naïve Bayes model. This global measure of CHR-related linguistic change was positively correlated with psychotic symptoms, including anhedonia ($R = 0.11$, $P = 6.1 \times 10^{-3}$), asociality ($R = 0.11$, $P = 6.7 \times 10^{-3}$), and disorganization ($R = 0.10$, $P = 1.6 \times 10^{-2}$). Conversely, CHR-specific language changes were negatively associated with social functioning ($R = -0.15$, $P = 1.6 \times 10^{-4}$) and with cognitive performance on verbal memory ($R = -0.11$, $P = 5.8 \times 10^{-3}$) and processing speed ($R = -0.14$, $P = 9.4 \times 10^{-4}$).

Conclusions: Subtle linguistic markers not only distinguish CHR individuals but also track with cortical abnormalities, clinical symptoms, and cognitive deficits. This supports the use of NLP-derived language features as scalable biomarkers of psychosis risk and provides insight into their potential neural underpinnings, suggesting potential clinical applications.

Keywords: Clinical high-risk for psychosis, Biomarker, natural language processing (NLP), neuroimaging

Disclosure: Nothing to disclose.

P97. EFFECT OF ENDOTOXIN ON ALCOHOL SELF-ADMINISTRATION AND PERIPHERAL CYTOKINES IN PEOPLE WITH AUD

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Background: Neuroimmune function is linked to negative reinforcement drinking and to the development of alcohol use disorder (AUD). Stress and chronic alcohol consumption have been found to increase microglial activity, a marker of neuroinflammation. Endotoxin is well-known to induce neuroinflammation and acute stress. In rodents, endotoxin increases levels of proinflammatory cytokines, an effect potentiated by alcohol. Endotoxin has led to long-lasting increases in voluntary alcohol intake in mice. In humans, those who binge drink or engage in heavy alcohol consumption have higher levels of proinflammatory cytokines, which are associated with higher alcohol craving and severity of alcohol problems. However, a provocation study to examine neuroinflammation-induced drinking has not been developed in humans and has direct clinical significance for the development and evaluation of pharmacotherapies targeting the neuroimmune system. We examined whether endotoxin (vs.

placebo) decreases time to initiate drinking and increases alcohol self-administration in people with AUD.

Methods: In this proof-of-concept study, we evaluated the effect of endotoxin (0.4ng/kg) on drinking behavior in $n=13$ women and men with current (past 6-months) DSM-5 AUD. Participants were randomly assigned to receive either endotoxin ($n=7$) or placebo ($n=6$) 1 h and 45 min before the start of a 2-h alcohol self-administration period. We used a well-validated human laboratory paradigm that models two critical features of drinking behavior: the ability to resist drinking (i.e., time to initiate drinking) and subsequent ad-libitum drinking. Participants were provided with a pre-determined amount of their preferred alcoholic beverage designed to raise blood alcohol levels to 0.12g/dL. Participants could consume as much or as little as they wished over the 2-h self-administration period. Peripheral cytokines (TNF α , IL-1 β , IL-6, IL-10) were collected throughout the self-administration period and participants provided self-reported ratings of alcohol craving, stress, and sickness symptoms.

Results: Our sample was comprised of 38.5% women, 61.5% men, 30.8% White, 38.5% Black, 15.4% Asian, and 15.4% Other. Participants were, on average, 39.0 years of age, consumed 34.5 drinks/week, scored 15.1 on the Alcohol Dependence Scale, and scored 18.2 on the Alcohol Use Disorders Identification Test. Pilot data suggests that individuals who drank in the endotoxin group consumed more alcohol than those who drank in the placebo group (87.7% vs. 60.1%, respectively; Cohen's $d=1.03$). Of those who consumed alcohol, individuals in the endotoxin group drank faster compared to those in the placebo group (52.0 minutes vs. 66.8 min, respectively; Cohen's $d=0.33$). TNF α , IL-1 β , and IL-6 increased from baseline starting at 2 h and 15 min following endotoxin administration and approximately 30 minutes into the 2-h self-administration period ($ps=0.03-0.05$). Alcohol craving peaked at 2 h and 45 min following endotoxin administration (visual analogue scale [VAS] = 78.0 vs. 64.8 in the endotoxin vs. placebo groups, respectively), although a drug $\times$ time interaction was not significant ($p=0.07$; Cohen's $d=0.46$). There was no difference in self-reported stress or sickness symptoms between the endotoxin and placebo groups ($ps>0.05$).

Conclusions: To our knowledge, this is the first examination of endotoxin, which directly induces neuroinflammation, on time to initiate drinking and subsequent alcohol self-administration. Our data suggests an important role for inflammation in alcohol consumption and peripheral cytokine-reactivity in people with AUD. Elucidation of neuroinflammatory mechanisms underlying drinking will ultimately inform potential treatment targets, such as neuroimmune modulators, for heavy alcohol use.

Funding: R03AA028361

Keywords: Inflammation and cytokines, Alcohol, Acute Stress, alcohol use disorder

Disclosure: Nothing to disclose.

P98. CHROMATIN ACCESSIBILITY PATTERNS IN PERIPHERAL BLOOD MONONUCLEAR CELLS AS PREDICTORS OF OPIOID USE DISORDER

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Background: Opioid use disorder (OUD) is a major public health crisis in North America, affecting over 6 million individuals aged 12 or older. In 2024 alone, opioid misuse was responsible for more than 50,000 deaths and contributed to an estimated annual economic burden exceeding \$78.5 billion dollars. Chronic opioid use is associated with transcriptomic and epigenetic brain

changes that drive craving and relapse, perpetuating OUD. Currently, no technique can longitudinally monitor brain epigenetic mechanisms across drug addiction phases or treatment responses in humans. However, evidence suggests a connection between blood-based epigenetic mechanisms and brain changes. Epigenetic modifications in blood have been found to mirror alterations in the central nervous system including those seen in neuropsychiatric disorders, suggesting the potential of blood-based epigenetic markers as indicators of brain-related conditions.

Methods: We conducted an assay for transposase-accessible chromatin with sequencing (ATAC-seq) and RNA sequencing (RNA-seq) on peripheral blood mononuclear cells (PBMCs) from 233 controls and 377 opioid-dependent individuals, identified per DSM-IV. All opioid-dependent subjects were receiving either buprenorphine or methadone maintenance therapy. Samples were sourced from the NIDA Center for Genetic Studies. Sequencing cohorts were matched for sex (Male: 63%; Female 37%), age, and race (70% White, 14% African American; and 16% other). Smokescreen genetic data was available for 90% of subjects and was used for Network Based Heterogeneity Clustering Analysis (NHC) to identify genes with single nucleotide polymorphisms over- and under-represented in the opioid dependent population. To predict opioid dependence diagnosis based on the ATAC-seq data, we applied various machine learning algorithms with 80% of samples used for training and 20% holdout set. Within the training set, we performed a 5-fold cross validation.

Results: Our analysis reveals significant alterations in chromatin accessibility and gene expression within PBMCs of opioid-dependent individuals, indicating dysregulation of inflammatory and immune signaling pathways. We detected 618 differentially accessible chromatin regions (DARs) and 817 differentially expressed genes (DEGs) at $FDR \leq 0.05$. DARs and DEGs were enriched for pathways related to extracellular matrix organization, cell adhesion, and immune system action. 62% of DARs correlate with the gene expression of the corresponding gene. These molecular signatures are being further examined in regard to their relationship with clinical measures of opioid use and treatment response, including diagnosis, addiction severity, Clinical Opiate Withdrawal Scale scores, and treatment dropout. The NHC analysis of the Smokescreen data identified several single nucleotide polymorphisms potentially associated with opioid dependence that reflect similar biological pathways and ontologies identified in the ATAC and RNA sequencing data. Finally, binary classification of opioid dependence using Bayesian Additive Regression Trees achieved robust performance (ROC AUC: 0.932 Accuracy: 0.876).

Conclusions: Differentially expressed genes and accessible genomic regions in PBMCs implicate pathways related to immune system function, cell adhesion, and extracellular matrix organization. Together with genetic findings, these results highlight specific biological systems as potential contributors to the pathophysiology of OUD. Moreover, machine learning models trained on peripheral chromatin accessibility profiles achieved high predictive accuracy, underscoring their potential as minimally invasive diagnostic tools for opioid dependence. Collectively, this work supports the development of blood-based biomarkers to advance precision medicine approaches for substance use disorders.

Keywords: Opioid addiction, Blood biomarker, Epigenetic, Machine learning, PBMC

Disclosure: Nothing to disclose.

P99. ASPARTATE TO ALANINE TRANSAMINASE (AST/ALT) BUT NOT LIVER FUNCTIONING DISCRIMINATES PARKINSON'S FROM OTHER DISEASES

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Background: Although 1-million Americans have Parkinson's disease (PD), patients are typically diagnosed at advanced stages when motor symptoms are apparent and substantial CNS damage has occurred. Identification of patients in prodromal, asymptomatic stages affords an opportunity to intervene with pharmaceuticals or other therapeutics to forestall clinical progression. Diagnosis currently depends on expert examination to which geographic access is limited. Simple, affordable, and rapid methods for detecting prodromal PD are essential as the global burden of PD is expected to increase.

A recent analysis of U.K. Biobank data identified aspartate to alanine transaminase (AST/ALT) among 13 blood analytes as a novel marker of PD risk. Elevations in AST/ALT were noted an average of 7.9 years prior to PD diagnoses. In an earlier study, AST was high up to 10 years prior to motor symptoms. Separate work reported negative correlations between AST/ALT and grip strength and walking speed, impaired in PD. In those already diagnosed, we hypothesized that high serum AST/ALT but not impaired liver function—determined by FIB4, a non-invasive scoring system to estimate liver disease—would discriminate PD from other diseases including alcoholic or other/unspecified cirrhosis, mild cognitive impairment (MCI), and Alzheimer's disease (AD).

Methods: Three independent, cross-sectional datasets were evaluated for relations between current diagnoses (ICD10 codes) and recent clinical laboratory tests including AST/ALT and FIB4 (age*AST/platelet count* $\sqrt{\text{ALT}}$), a serum estimate of liver disease. The first dataset comprised 712 participants from our laboratory [lab: 39 PD, 45 MCI, 240 Alcohol Use Disorder (AUD), 101 HIV, 100 HIV + AUD, 187 healthy controls] that had upper motor composite scores based on grooved pegboard, fine finger movement, or finger tapping performance. The second, derived from the Stanford Research Repository (STARR), included 1,277 individuals [188 PD, 27 MCI, 25 Vascular dementia (VD), 106 AD, 119 alcoholic cirrhosis, 48 hepatic fibrosis, 764 hepatic cirrhosis]. The third, originating with the Medical Information Mart for Intensive Care (MIMIC)-IV (<https://physionet.org/content/mimiciv/3.1/>), incorporated data from 6805 individuals (827 PD, 527 VD, 833 AD, 2266 alcoholic cirrhosis, 671 hepatic fibrosis, 1681 hepatic cirrhosis). Welch's tests for unequal variance were used to evaluate AST/ALT and FIB4 levels among the various diagnoses; a simple regression evaluated the relation between AST/ALT and motor performance scores.

Results: Our sample demonstrated that AST/ALT is high in PD relative to the other 5 groups ($F = 3.18$, $p = 0.009$), whereas FIB4 was high in HIV + AUD relative to the remaining groups ($F = 4.50$, $p = 0.0013$). In the STARR data, AST/ALT was high in PD and alcoholic cirrhosis relative to the other diagnoses ($F = 7.96$, $p < 0.0001$), but FIB4 was only high in cirrhosis (both alcoholic and other/unspecified; $F = 51.19$, $p < 0.0001$). In the MIMIC dataset, AST/ALT distinguished PD and alcoholic cirrhosis ($F = 174.05$, $p < 0.0001$), while FIB4 isolated alcoholic cirrhosis ($F = 106.27$, $p < 0.0001$) from the other groups. Our sample further demonstrated a correlation between high AST/ALT and worse upper motor performance ($r = -11$, $p = 0.0065$).

Conclusions: The results of analyzing of 3 unique datasets converge to confirm that PD can be differentially diagnosed using a combination of AST/ALT and FIB4: AST/ALT but not FIB4 is high in PD relative to other diagnoses including MCI, AD, VD, HIV, AUD, and liver disease. Together, these results suggest that high AST/ALT does not reflect liver disease. Instead, especially in the context of PD, a muscle source of these enzymes should be considered. Prospective longitudinal studies to predict a PD diagnosis using a combination of AST/ALT and FIB4, and preclinical studies to better understand non-hepatic functions of AST and ALT are thus warranted.

Keywords: serum levels, motor function, Alcoholic Liver Disease

Disclosure: Nothing to disclose.

P100. PROSPECTIVE REPLICATION OF AN EEG BIOMARKER FOR PREDICTING CHANGES IN ATTENTION IN ALTO-203, AN H3 INVERSE AGONIST

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Background: Neuropsychiatric disorders are markedly heterogeneous, which leads to pharmacotherapies developed in broad populations failing or demonstrating minimal effect sizes over placebo. The identification of likely responders through objective markers may enhance treatment effects by targeting subpopulations for a given drug. ALTO-203 is an H3 inverse agonist that enhances the release of dopamine, norepinephrine, acetylcholine, and histamine, suggesting a pro-wakefulness and pro-cognitive profile that may be beneficial for indications such as Major Depressive Disorder (MDD). A prior Phase 1 trial in healthy volunteers demonstrated significant acute benefits of ALTO-203 on subjective positive emotion and alertness, along with improvements in sustained attention and reduced theta/beta ratio, a neurophysiological index of cortical arousal and attentional control. Moreover, subjects with higher theta/beta ratios at baseline showed greater changes in sustained attention with ALTO-203 than subjects with lower theta/beta ratios, suggesting that ALTO-203 may be preferable for treating attentional deficits in individuals with higher theta/beta ratios. The goal of the current study was to replicate the findings from the Phase 1 trial in a cohort of individuals with MDD.

Methods: A total of 69 participants with MDD and anhedonia (defined as a Snaith Hamilton Pleasure Scale [SHAPS] > 28) were enrolled in a Phase 2 proof-of-concept (POC) trial of ALTO-203 (NCT06391593). Participants underwent a single-dose period, in which they received two single-doses of ALTO-203 (25 μg and 75 μg) and placebo in a randomized, three-way crossover design with an at least 10-day washout period. The primary outcome was an acute change in positive emotion assessed by the Bond-Lader Visual Analog Scale (BL-VAS). Participants underwent cognitive assessments, electroencephalography (EEG), and sleep and activity monitoring using a wearable device. Following the single-dose period, participants were randomized to receive daily administration of ALTO-203 (25 μg or 75 μg QD) or placebo over 28 days to evaluate extended safety, tolerability and pharmacokinetics. Here, our analysis set includes 52 participants that passed blinded prospective central case review and completed the single-dose period. Mixed-effects models were used to evaluate the efficacy of ALTO-203 versus placebo on outcome measures; subgroup analyses of subjects with higher theta/beta ratios were also tested via mixed-effects models to further replicate findings from the Phase 1 trial. Statistical significance was evaluated with two-sided p -values with no correction for multiple comparisons.

Results: There was no significant separation between ALTO-203 and placebo on the BL-VAS due to a higher-than-expected placebo response ($p > 0.407$). Compared to placebo, ALTO-203 25 μg significantly improved sustained attention (25 μg $p < 0.05$; 75 μg $p = 0.06$) and decreased theta/beta ratios (25 μg $p < 0.05$). Improvement in sustained attention with ALTO-203 was greatest among participants with high baseline theta/beta ratios (25 μg : $p < 0.01$; 75 μg : $p < 0.05$ vs. placebo). Increased wakefulness (as measured by reduced minutes asleep) as measured by wearable devices supported the wake-promoting effects of ALTO-203 (25 μg : $p < 0.05$; 75 μg : $p < 0.001$ vs. placebo). ALTO-203 displayed predictable accumulation over multiple doses and was well tolerated, with insomnia as the most frequent adverse event – consistent with its wake-promoting profile.

Conclusions: These data replicate prior findings that ALTO-203, a novel H3 inverse agonist, improves sustained attention and cortical arousal, and that individuals with higher theta/beta ratios are more likely to realize the greatest benefit in sustained attention. These data suggest that ALTO-203 may be best suited for disorders with hypersomnolence or excessive daytime sleepiness, along with deficits in sustained attention, or attention deficit hyperactivity disorder with partial response to stimulants.

Keywords: Biomarker Prediction, precision psychiatry, Cognition

Disclosure: Alto Neuroscience, Employee, Self.

P101. EFFECTS OF SLEEP QUALITY AND PHYSICAL ACTIVITY ON DEPRESSION AND STRESS ACROSS THE ADULT LIFESPAN: FINDINGS FROM THE ADULT AGING BRAIN CONNECTOME (AABC) CONSORTIUM

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Background: Sleep and physical activity can impact mood in older adults, potentially acting as a buffer against cognitive decline. We aimed to examine the relationship between physical activity, sleep and depression in a cross-sectional study of a large sample of adults (N = 180; mean age: 66.6 (SD = 13.4), 40–95 years) from the Human Connectome Project – Aging (HCP-A) and the Adult Aging Brain Connectome (AABC) studies.

Methods: Participants completed the International Physical Activity Questionnaire (IPAQ), Pittsburgh Sleep Quality Index (PSQI), Ongoing Chronic Stressors Scale (OCSS), and Center for Epidemiological Studies – Depression (CES-D) scales. Physical activity was measured in units of the metabolic equivalent of task (MET). We used general linear models to examine how the PSQI and IPAQ were related to OCSS and CES-D, controlling for age, sex, ethnicity, and study site. Age was treated as a factor with 2 levels (<65 and ≥65 years).

Results: We found that both physical activity and sleep were associated with depression and stress differentially across age groups. For depression, both the age group-by-physical activity ($F(1, 156) = 5.41, p = 0.02$) and age group-by-sleep quality ($F(1, 156) = 4.29, p = 0.04$) interaction terms were significant. For stress, the age group-by-physical activity ($F(1, 156) = 6.09, p = 0.01$) was significant while the age group-by-sleep quality ($F(1, 156) = 3.16, p = 0.07$) term did not reach significance. Specifically, physical activity and sleep quality were more strongly related to depression in the younger (IPAQ: $\beta = -2.3, p = 0.001$; PSQI: $\beta = 1.3, p < 0.0001$) than older group (IPAQ: $\beta = 0.2, p = 0.8$; PSQI: $\beta = 0.6, p = 0.002$). Similar findings were observed for stress: younger group (IPAQ: $\beta = -1.3, p = 0.0007$; PSQI: $\beta = 0.3, p = 0.01$) showed significant associations while the older group (IPAQ: $\beta = -0.1, p = 0.7$; PSQI: $\beta = 0.03, p = 0.7$) did not.

Conclusions: Results from this large sample illustrate an age-related difference in how physical activity and sleep quality affect mental health. Worse sleep quality and lower physical activity were associated with greater depression and more stress in the younger group (<65 years). Future analyses will determine potential protective factors of sleep quality and physical activity for cognitive decline.

Keywords: Depression, Sleep disturbances, Chronic social and non-social stress, Aging; Cognition; Stress; Acetylcholinesterase; Splice Variants

Disclosure: Nothing to disclose.

P102. FLUID MOLECULAR BIOMARKER DISCOVERY IN A WELL-PHENOTYPED LONGITUDINAL COHORT OF PSYCHOSIS SPECTRUM DISORDER

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Background: For psychosis spectrum disorders (schizophrenia to bipolar 1 disorder), significant within-diagnosis heterogeneity and transdiagnostic sharing of genetic risks and symptoms create serious obstacles to patient selection for clinical trials and other case-control studies. We present the Psychiatric Biomarkers Network (PBN), a multisite, multisector (academia and industry) effort, modeled on the Alzheimer's Disease Neuroimaging Initiative (ADNI) and focused transdiagnostically on psychosis spectrum disorders. Recent efforts from ADNI demonstrate that blood analytes selected based on prior neuroimaging and CSF analyses can predict clinical progression, supporting the overall PBN strategy of first identifying candidate biomarkers from longitudinal CSF analysis. Cognitive deficits are variable but present in a majority of psychosis patients, are highly predictive of functional outcomes, and remain unaddressed by existing therapeutics. Thus biomarkers associated with cognitive phenotypes are a central focus of our efforts. The goal of the PBN is to accelerate therapeutic development for psychosis spectrum disorders by providing industry with robust, reproducible and objective biomarkers that enable patient selection, prognosis, and longitudinal tracking of the biological mechanisms underlying symptoms and functional deficits.

Methods: The PBN is an ongoing prospective, longitudinal fluid biomarker study collecting paired cerebrospinal fluid (CSF) and blood samples from patients and controls along with clinical, neuroimaging, and cognitive phenotypes. A rigorous protocol has been implemented across four clinical sites (Mt Sinai, Penn, Yale, Northwell) and coordinated by the Broad. We enroll patients aged 18–55 who are within ten years of an initial diagnosis of schizophrenia, schizoaffective disorder, schizophreniform disorder, or bipolar 1 disorder, along with demographically matched control subjects. The Penn Computerized Neurocognitive Battery (CNB) is administered providing measures on 14 tests evaluating five domains: Executive, Memory, Complex Cognition, Social Cognition and Sensorimotor Speed. Assessment of symptom severity and functioning is conducted concurrently along with structural and resting-state functional neuroimaging, CSF and blood sampling. Biofluid collection, processing, banking and distribution is managed by the Indiana University Genetics Biobank. Targeted molecular analysis of biofluid samples is hypothesis-driven, initially focused on analytes hypothesized to be associated with excessive synapse elimination (e.g., complement factors, synaptic proteins, and inflammatory markers). Complementary hypothesis-generating analyses are based on use and comparison of unbiased proteomics platforms (Somascan, Olink, Nulisa). Participants were followed longitudinally in a repeated measures design.

Results: In its first four years of operation at three clinical sites (with a fourth site beginning enrollment in mid-2025) the PBN performed or scheduled baseline visits including successful CSF collection for 118 participants (69 patients, 49 controls). Retention rates are approximately 70% for each longitudinal follow-up visit (semi-annual during the pilot phase; annual biosample collection with semi-annual clinical and cognitive phenotyping under the current protocol), with some of the earliest enrolled participants

returning for as many as eight separate lumbar punctures. Symptom and cognitive data from the pilot phase were analyzed for the first 77 participants (44 cases, 33 Controls) with similar demographics. The groups differed significantly in severity of general, positive and negative symptoms across three 6-months intervals ($F = 83.3$, $F = 54.5$, $F = 57.2$, $p < 0.0001$) with patients exhibiting poorer functioning ($F = 30.9$, $p < 0.0001$). CNB results showed consistent and, importantly for biomarkers, stable deficits in patients, most pronounced for executive functioning and social cognition for accuracy and speed ($p < 0.001$), and in episodic memory accuracy ($p < 0.001$). The cognitive deficits were correlated with symptom severity and functional outcome. Preliminary molecular results, presented separately in the poster by Triana-Baltzer et al., are consistent with the hypothesis that fluid biomarkers will enable a biological definition of subsets of psychosis spectrum patients.

Conclusions: Patients with diagnoses of schizophrenia spectrum disorders or bipolar 1 disorder, as well as demographically matched control subjects, can be successfully recruited and retained for longitudinal CSF collection paired with blood sampling and extensive phenotyping. Patient and caregiver outreach and education are key to recruitment success, and retention benefits from regular contact with the study team. Careful planning, training and execution yields robust clinical and cognitive phenotype assessments across clinical sites, with stable measurements and expected group differences. Preliminary molecular data, presented in detail in our companion poster, are consistent with the potential for fluid biomarkers to advance psychiatric drug development by enabling prognosis and patient stratification.

Keywords: CSF Biomarkers, psychosis spectrum disorders, Cognitive biomarkers, schizophrenia and bipolar disorders, longitudinal study

Disclosure: Nothing to disclose.

P103. RNA METRICS FOR PRECISION IMMUNOPSYCHIATRY IN MAJOR DEPRESSION

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Background: Systemic inflammation contributes to some cases of Major Depressive Disorder (MDD) and provides a target for intervention if inflammatory etiology can be accurately identified. Most precision immunopsychiatry research has used C-reactive Protein (CRP) to mark inflammation, but that biomarker may miss activity other pro-inflammatory signaling pathways besides the IL6/STAT3 pathway that drives CRP. Recent research implicated the NF-kappaB, AP-1 and Interferon Response Factor (IRF) immunoregulatory pathways in MDD (Savitz et al., *Brain, Behavior and Immunity*, 2025; PMID: 39532200). The present analyses tested whether bioinformatic measures of those pathways' activity based on circulating leukocyte RNA profiles might supplement or outperform CRP in identifying MDD cases with an inflammatory component.

Methods: We performed new secondary bioinformatic pathway analyses of genome-wide RNA profiles collected from peripheral blood mononuclear cells (PBMC) from 159 individuals (79 MDD, 80 healthy control HC; Le et al., *Translational Psychiatry*, 2018; PMID: 30185774). We applied machine learning algorithms to pre-specified sets of 19 gene transcripts involved in the NF-kappaB and AP-1 canonical inflammatory pathways and 34 gene transcripts involved in the Type I interferon gene regulation pathway involving IRF transcription factors. Resulting scores

indexing the canonical inflammatory pathway (INFLAM) and Type I interferon pathway (IFN) were tested for association with MDD case vs HC control status in logistic regression models that controlled for age (median 28 years, range 18–55), sex (67 females, 59 males), and RNA assay batch, as well as log plasma CRP concentrations where indicated.

Results: In a new bioinformatic analysis of RNA data from a previously published case-control study of 159 individuals (79 MDD, 80 HC), CRP > 3 mg/L marked 34% of 79 total SCID-I/NP-confirmed MDD cases (OR = 1.64, biserial $r = 0.13$, kappa = 10% accuracy above chance, $p = 0.168$) implying 66% of MDD cases to be “non-inflammatory.” To determine whether other pro-inflammatory signaling pathways might remain undetected in that low-CRP fraction, we quantified pre-specified sets of mRNA transcripts marking activation the NF-kappaB/AP-1 (INFLAM) and Type I interferon/IRF (IFN) signaling pathways in PBMC. 88% of MDD cases showed elevated INFLAM RNA (>8.5 log2-TPM; OR = 3.91, $r = 0.35$, kappa = .22, $p = 0.001$), resulting in 2.6-fold increased detection of putatively inflammatory MDD cases compared to CRP. All IFN+ individuals were also INFLAM+. However, those who scored positively for IFN+ in addition to INFLAM+ showed substantially enhanced MDD prevalence relative to IFN-/INFLAM+ (83% vs. 50%, $p = 0.003$). Differential MDD prediction remained robust in logistic regressions that controlled for age, sex, and BMI (CRP + OR = 1.15, $p = 0.733$; INFLAM + OR = 4.70, $p < 0.001$; IFN + /INFLAM + OR = 15.36, $p < 0.001$). RNA metrics also predicted MDD in analyses controlling for CRP (INFLAM +: OR = 5.22, $p < 0.001$; INFLAM + /IFN +: OR = 16.84, $p < 0.001$), whereas CRP showed no significant prediction of MDD beyond that available from INFLAM and IFN RNA metrics (OR = 1.23, $p = 0.215$).

Conclusions: MDD precision psychiatry may benefit from supplementing plasma measures of systemic inflammation (e.g., CRP) with cellular RNA measures that capture additional pro-inflammatory signaling pathways (NF-kB, AP-1, IRF) that are missed by CRP. Current analyses relying on CRP alone likely miss a large fraction of MDD cases that involve elevated inflammatory signaling. RNA-based bioinformatic measures of NF-kappaB/AP-1 and IRF activity substantially outperform CRP in predicting MDD, and represent promising biomarkers for improving the precision of case targeting and intervention selection in immunopsychiatry.

Keywords: immunopsychiatry, inflammation, C-Reactive Protein, NF-kappaB, Interferon Response Factors

Disclosure: Nothing to disclose.

P104. DEMONSTRATING THE POTENTIAL OF UNTARGETED HAIR PROTEOMICS FOR PERSONALIZED BIOMARKERS

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Background: Biomarker research in psychopathology increasingly employs high-dimensional omics approaches. Yet, proteomics based on human hair remain largely unexplored, despite its potential to efficiently capture stable biological signals accumulated over weeks to months. This study leveraged machine learning to investigate the potential of the hair proteome—all detectable peptides and proteins—as a biomarker source for stress-associated psychopathology.

Methods: We analyzed protein profiles from hair segments of women with non-suicidal self-injury disorder and healthy controls (N = 68) using Mass Spectrometry. Group membership was

predicted using cross-validated Partial Least Squares Discriminant Analysis (PLS-DA). Prediction robustness was assessed using different data imputation, participant-subsampling, and variable perturbation strategies. Further, the intrinsic proteomic structure was assessed with two non-supervised clustering algorithms. Proteome-wide group differences in single proteins were assessed using t-tests and FDR correction ($q = 0.05$).

Results: Of 1114 identified proteins, 611 were sufficiently abundant for analyses. PLS-DA achieved 84.4% cross-validated accuracy for classification of clinical groups ($p < 0.001$). This accuracy remained robust in sensitivity analyses and outperforms models based on data-derived clusters (60%), stress-related proteins (73%), and simulated hair cortisol from meta-analytic effect sizes (53–59%). Cross-validated predicted class probabilities strongly correlated with clinical symptoms and well-being ($r > 0.60$). Key predictive proteins were linked to pain perception, oxidative stress, and cholesterol homeostasis. Approximately 15% of proteins differed significantly between groups, with the strongest candidates related to ribosomal function—an emerging target in depression.

Conclusions: Hair proteomics offers a novel, non-invasive tool for identifying stable, biologically grounded markers of psychopathology. Unlike blood or saliva, hair provides a time-resolved molecular archive, enabling retrospective insight into biological processes accumulated over weeks to months—opening new possibilities for early risk detection and personalized psychiatric care when moving beyond single compounds.

Keywords: Proteomics, Mental Disorders, Biomarker

Disclosure: Nothing to disclose.

P105. BRAIN PROTEOME OF WELL-BEING: A PROTEOME-ASSOCIATION STUDY UNVEILS GENETICALLY-REGULATED PROTEINS AND POTENTIAL DRUG TARGETS RELATED TO WELL-BEING

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Background: Human genetic evidence has been reported as a proxy for the success of drugs in clinical trials due to its ability to pinpoint causal mechanistic links between drug targets and diseases, with many drugs approved by the FDA supported by human genetic evidence associating the intended pharmacological target with the indication. Proteins are common drug targets; thus, the identification of proteins with strong evidence that their genetic variants influence their abundance or expression in a given condition can support drug development efforts. This can be assessed by Protein quantitative trait locus (eQTL), which connects genetic variants to protein levels, identifying proteins that are in the mediation path from genotype to phenotype. Well-being, a multidimensional construct encompassing subjective, psychological, and social functioning, has been increasingly recognized as an important component of mental health, with the mere absence of a psychiatric disorder not fully equating to optimal mental health. We aim to reliably identify proteins causally related to well-being by conducting a protein-wide association study (PWAS) supported by convergent evidence using transcriptome-wide association study (TWAS), Mendelian randomization, and colocalization. By employing such approach, we expect to identify proteins with a strong genetic basis, from genes to transcripts to proteins with evidence of causality in influencing well-being.

Methods: We obtained four genome-wide association study (GWAS) summary statistics on well-being: subjective well-being ($n = 563,176$), well-being spectrum ($n = 2,370,390$), positive

affective ($n = 410,603$), and life satisfaction ($n = 80,852$). For proteomics, we used as reference the postmortem human dorsoprefrontal cortex (dPFC) brain proteomes from ROSMAP ($n = 12,691$ proteins) and Banner ($n = 11,518$ proteins), as the datasets for the discovery and replication PWAS, respectively, and their protein quantitative trait locus (pQTL). For TWAS, we employed precomputed mRNA weights from brain samples provided by ROSMAP, Mayo, and Mount Sinai Brain Bank studies ($n = 6818$ cis variants transcripts), and from participants of the Common Mind Consortium. Both PWAS and TWAS were done using FUSION. After, we performed Mendelian randomization (MR) for all proteins found significant in the discovery PWAS, followed by colocalization in those with a significant MR. Finally, we verified the cell-type and tissue-specificity of those proteins and their gene ontology.

Results: 118 unique proteins were identified as related to well-being in the discovery PWAS, with 56 also significant with the same direction in the replication PWAS. In order to strengthen our confidence that the proteins discovered in the PWAS were indeed causally related to well-being, we conduct two TWAS to verify if the genes of the proteins related to well-being identified in the discovery PWAS also had their transcripts associated with well-being. 41 proteins were replicated in at least one TWAS. Twenty-two proteins were found altered with the same direction in the replication PWAS and had their transcripts altered with the same direction as in the discovery PWAS in the TWAS (B3GALT1, CDH13, CSDC2, CSE1L, GMPPB, GPX1, LRP4, MGST1, MMAB, PLEKHB1, SHMT1, PACSIN3, SLC25A12, SLC5A6, ATG7, DARS2, MADD, PRKCD, SLC30A9, ALDH16A1, TREX1, VKORC1). Eleven proteins had evidence from both MR and colocalization (mapped to genes CDH13, CSD2, CSE1L, GMPPB, ICA1L, P2RX7, PACSIN3, RAB27B, SLC25A12, TMEM106B, RGS6). The differently expressed genes were downregulated in the amygdala, hippocampus, and putamen, and both up- and down-regulated in the substantia nigra and caudate nucleus (FDR $q < 0.05$). The 11 proteins associated with the genes with evidence from both MR and colocalization were enriched or depleted in excitatory neurons (ICA1L, P2RX7, RGS6, and SLC25A12), inhibitory neurons only (CSE1L and TMEM106B), and in both excitatory and inhibitory neurons (CDH13, RGS6, and SLC25A12). P2RX7 was enriched in oligodendrocytes in addition to excitatory neurons, and PACSIN3 was found enriched only in oligodendrocytes. None was enriched in astrocytes or microglia. The replicated proteins were associated with several biological pathways, notably vitamin metabolic process and biosynthesis of cofactors, particularly cobalamin, pantothenic acid, pyridoxine, biotin, and niacin, which contribute to the synthesis of methionine, thymidylate, purines, and coenzyme A (CoA). Also, the response to hydroperoxide was found enriched.

Conclusions: We identified 11 genetically-regulated proteins related to well-being with both gene and transcript evidence and reinforced their causal relationship with well-being with convergent support from MR and colocalization. These proteins were found enriched or depleted in several brain regions and cell types, particularly neurons, and are implicated in a myriad of biological processes, such as the synthesis of methionine, purines, and CoA, all vital for cellular energy production, and the production of reactive oxygen species, influencing oxidative damage, mitochondrial function, and inflammatory responses. If validated in subsequent experiments, these identified proteins could lead to the development of precision therapeutics, potentially optimizing well-being by targeting individuals with different levels of protein expression with different drugs, with the potential of ultimately improving mental health at scale.

Keywords: Translational biomarker approaches to drug development, Precision psychiatry, Drug Discovery - new approaches, Novel Targets, Precision mental health

Disclosure: Nothing to disclose.

P106. HIGHER PULSE WAVE VELOCITY IS ASSOCIATED WITH LOWER GLOBAL AND REGIONAL BRAIN VOLUME IN YOUTH WITH AND WITHOUT BIPOLAR DISORDER: EVIDENCE FOR SEX-SPECIFIC EFFECTS

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Background: Background: Substantial evidence highlights the importance of heart-brain connections in adults. Arterial stiffness, indexed by non-invasive pulse wave velocity (PWV), provides predictive value for cardiovascular and cerebrovascular events beyond that of traditional cardiovascular risk factors (CVRFs). Furthermore, PWV is associated with lower brain volume and poorer cognition in adults. Whether similar associations exist in youth is unknown. Prior work in adolescents shows that elevated CVRFs, even within normative ranges, are adversely associated with brain structure and function. Given that neurodevelopment during youth is characterized by heightened plasticity and vulnerability, understanding the link between arterial stiffness and brain health in adolescents could inform preventative interventions and confer long-term benefits. Here we present preliminary findings from a controlled study focused on youth with bipolar disorder (BD), who are at increased risk of early-onset cardiovascular disease and accelerated atherosclerosis.

Methods: Methods: Sixty-seven youth, between 13 and 20 years of age ($n = 20$ BD, $n = 47$ controls) have participated to date. All participants completed the Kiddie-Schedule for Affective Disorders and Schizophrenia for School-Age Children- Present and Lifetime Version (K-SADS-PL) semi-structured interview to ascertain psychiatric diagnoses, clinical characteristics, and treatments. We measured aortic PWV using magnetic resonance imaging, specifically cardiac gated cine images and two-dimensional phase contrast images above the aortic valve and iliac bifurcation. Additionally, we measured brain structure (volume, thickness and surface area) using T1-weighted brain imaging. We used general linear models to test the association of PWV with cerebral gray matter volume and hippocampal volume, controlling for age, sex, and body mass index. Finally, we used cortical vertex-wise analysis to examine the association of PWV with cortical structure. Sex differences were evaluated in sex-stratified and sex-interaction analyses.

Results: Results: In the full sample, Higher PWV was associated with lower total gray matter volume ($\beta = -0.30$, $p = 0.04$) and hippocampal volume ($\beta = -0.46$, $p = 0.001$). In sex-stratified analyses, higher PWV was associated with lower hippocampal volume in males ($\beta = -0.45$, $p = 0.01$) but not females ($\beta = -0.24$, $p = 0.18$). We did not observe any significant within-sex associations between PWV and cerebral gray matter volume ($p > 0.1$). In vertex-wise analyses, higher PWV was associated with lower area in the banks of the superior temporal sulcus ($p = 0.03$). Additionally, we observed a significant sex-by-diagnosis interaction whereby higher PWV was associated with lower thickness in the superior frontal gyrus to a significantly greater extent in males vs. females ($p = 0.04$). Analyses are ongoing and a sample of 85 is anticipated by the time of presentation.

Conclusions: Conclusion: This study provides preliminary evidence that higher arterial stiffness, as indexed by PWV, is associated with lower cerebral and regional brain volumes in youth. Findings also reveal sex-specific effects, with stronger associations between PWV and hippocampal volume and frontal cortical thickness in males. These findings extend the growing body of evidence that heart-brain connections are evident in youth and suggest that PWV may serve as a potential target for preventive interventions. Notably, PWV-brain associations were independent of obesity, aligning with findings in adult

populations that PWV reflects vascular risk beyond traditional CVRFs. Our prior work showed that cerebrovascular pulsatility is elevated in youth with BD and can be reduced through exercise, supporting the potential for modifiable interventions from a cerebrovascular perspective. Future studies with larger, longitudinal samples that integrate neuroimaging, blood-based biomarkers, and mechanistic probes are warranted to clarify causal pathways and inform strategies to mitigate long-term adverse heart-brain connections.

Keywords: Bipolar Disorder, brain structure, cardiovascular function, arterial stiffness, Adolescence

Disclosure: Nothing to disclose.

P107. NEUROCHEMICAL PROFILE OF MANIA AND ASSOCIATION WITH SUBSEQUENT TREATMENT RESPONSE IN YOUNG ADULTS DURING A FIRST MANIC EPISODE: A PROTON MAGNETIC RESONANCE SPECTROSCOPY STUDY

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Background: The neurophysiology of bipolar disorder, particularly the acute manic state, remains poorly understood. Proton Magnetic Resonance Spectroscopy (MRS) allows measurement of multiple metabolites thought to underly important neural attributes implicated in bipolar disorder, including cellular signaling and membrane homeostasis (myoinositol, MI); neuronal integrity (N-acetyl aspartate, NAA); neuronal energy production (creatine, Cr); cell membrane activity and signal transduction (choline, Cho); and glutamatergic function (glutamate, Glu and glutamate/glutamine/GABA, Glx). This study compared MRS metabolites between adolescents/young adults during a first manic episode, before treatment initiation, and a healthy control group. Further, in the mania group, we assessed the association of these metabolites with subsequent response to treatment.

Methods: Adolescents and young adults with bipolar I disorder ($n = 53$, 30 F/23 M, mean age 18 ± 5) were recruited during their first manic episode, pseudorandomized to receive either lithium or quetiapine, and followed for eight weeks. 4T MRI scans including 1H-MRS were performed prior to treatment initiation and 1 and 8 weeks later. Healthy participants ($n = 54$, 30 F/24 M, mean age 22 ± 6) were scanned at the same intervals. MRS voxels were placed in the anterior cingulate cortex (ACC) and the left and right ventrolateral prefrontal cortices. At the 8 week mark, BD participants were classified as remitters ($YMRS \leq 10$) or nonremitters ($YMRS > 10$). The current study analyzed data from the first 1H-MRS scan. Mania and control group comparisons used general linear models to compare baseline MRS metabolites between groups, covarying for age. For remission analyses, remitter and non-remitter groups were compared using general linear models to compare baseline MRS metabolites between groups, covarying for age.

Results: In all analyses, the ACC showed more pronounced between-group differences than ventrolateral prefrontal cortex. At baseline, multiple ACC metabolites were significantly higher in the mania compared to control group: NAA, Cr, Co, and Glu (all $p < 0.02$). Of the 39 BD participants with available longitudinal data, 26 were in remission at 8 weeks. Baseline ACC Cho was significantly higher in non-remitters compared to remitters ($p = 0.036$).

Conclusions: Compared to control participants, participants with mania demonstrated a distinct neurochemical profile in the anterior cingulate cortex, with increased levels of multiple metabolites reflecting abnormalities in various cellular functions. Further, higher baseline level of choline in the mania group was associated with

subsequent non-response to treatment. This indicates that cell membrane activity and signal transduction abnormalities may be related to treatment response in bipolar disorder. Further analyses will characterize ability of neurochemical profile to predict treatment response, and changes in MRS metabolites over time in response to lithium or quetiapine treatment.

Keywords: bipolar disorder, mania, magnetic resonance spectroscopy

Disclosure: Johnson and Johnson, Contracted Research, Self.

P108. EXPLORING THE RELATIONSHIP BETWEEN INFLAMMATION AND BLOOD-BRAIN BARRIER FUNCTION IN BIPOLAR DISORDER TYPE I

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Background: The neutrophil-to-lymphocyte ratio (NLR) is a simple and clinically significant marker of peripheral inflammation, with elevated levels linked to various psychiatric disorders. However, existing literature presents mixed results. Increased neutrophil counts can cause the release of pro-inflammatory cytokines, potentially disrupting brain function and the blood-brain barrier (BBB), leading to negative outcomes. This study aimed to investigate the relationship between NLR and S100B, a peripheral marker of BBB disruption, alongside clinical features of bipolar disorder type I (BD-I) during acute episodes, shedding light on the inflammatory mechanisms underlying this mood disorder.

Methods: This pilot study provides preliminary data from an ongoing research project. We recruited 10 patients (aged 18–40) with BD-I, presenting acute manic or mixed symptoms, from Zucker Hillside Hospital, in New York. Informed consent was obtained, followed by clinical assessments via SCID for diagnosis and symptom evaluation using HAMD for depression, YMRS for mania, and BPRS for psychosis. The MATRICS Consensus Cognitive Battery (MCCB) was also administered, and eight patients underwent magnetic resonance imaging (MRI) to explore neurobiological correlates, including DTI-derived free water.

Results: NLR values ranged from 1.6 to 3.8 (Mean = 2.66, SD = 0.78). A higher NLR correlated with more severe depressive (Pearson $R = 0.77$, $p = 0.01$) and psychotic symptoms (Pearson $R = 0.66$, $p = 0.04$), but not manic symptoms. S100B did not correlate with NLR, mood, or cognitive symptoms, but was positively associated with increased free water in specific white matter tracts (e.g., anterior corona radiata, $p = 0.006$).

Conclusions: Both NLR and S100B associate with key clinical features in BD-I patients. NLR reflects acute inflammation related to higher depressive and psychotic symptoms, while S100B indicates BBB disruption linked to white matter abnormalities. The lack of correlation between NLR and S100B suggests that NLR may represent acute stress, whereas S100B may highlight brain dysfunction in BD-I.

Keywords: Blood-Brain-Barrier, Mood and Cognition, Inflammation, Bipolar Disorder

Disclosure: Nothing to disclose.

P109. ACUTE THC NORMALIZED DEFICIENT TEMPORAL PERCEPTION OF PEOPLE WITH BIPOLAR DISORDER

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Background: Altered temporal perception (ability to perceive time accurately and precisely), can negatively impact behavior and cognition. Poorer performance on temporal estimation tasks has been associated with both mania and depression in people with bipolar disorder (BD), thus deficient temporal perception may be a trait of BD. People with BD report using cannabis to ameliorate cognitive symptoms, including those cognitive functions regulated in-part by temporal perception. Acute cannabis use drove slowed temporal perception in healthy adults and rodents. Previously, we reported that people with BD who do not use cannabis had impaired timing precision, whereas those who use cannabis exhibited similar timing precision to healthy comparison participants (HC) measured using the cross-species temporal discrimination task (TDT). Participants with BD tended to over-estimate short intervals more often than healthy comparison participants. To expand upon these findings, we tested the impact of acute $\Delta 9$ -tetrahydrocannabinol (THC) on people with BD and HC who do not use cannabis. We hypothesized that within the placebo group, BD participants would have worse TDT performance relative to the HC participants, but that acute THC administration in BD participants would be equivalent TDT performance to the HC participants administered placebo.

Methods: We recruited 17 participants with BD and 10 HC that do not regularly use cannabis (<4x/month). Participants were randomized into one of two treatments (CT ID: NCT04231643); 5 mg of dronabinol (THC) or placebo (in capsule). Two hours post-drug ingestion, participants completed the TDT, where participants had to judge whether time intervals (ranging from 1.25 to 4.25 s) were shorter or longer than a criterion interval (2.75 s). Psychometric curves were generated to test for group differences in perceived interval duration between the groups. Planned comparison t-tests were conducted based on our a priori hypotheses (i.e., HC +placebo vs. BD +placebo, BD +placebo vs. BD + THC) to test for differences in TDT performance between the four comparison groups; HC+placebo ($n = 6$), HC + THC ($n = 4$), BD +placebo ($n = 9$) and BD + THC ($n = 8$). Primary outcome measures included Weber Fraction (WF; timing precision) and T50 (timing accuracy). Difference Limen (DL), a secondary measure of timing precision not normalized to the stimulus magnitude, was also calculated. Effects of gender and age on primary outcome measures were tested.

Results: There were no significant differences in gender distribution or age between the four comparison groups. The BD+placebo group had significantly lower T50 compared to the HC+placebo ($t(12) = 2.92$, $p = 0.013$, $d = 1.6$) and BD + THC group ($t(13) = 2.47$, $p = 0.028$, $d = 1.28$). There were no significant differences in WF between groups. DL was lower in the BD +placebo group compared to the HC+placebo group ($t(12) = 2.18$, $p = 0.05$, $d = 1.18$) and there was a trend of lower DL in the BD +placebo group compared to the BD + THC group ($t(13) = 1.91$, $p = 0.079$, $d = 0.99$). Psychometric curves for TDT performance showed that there was a leftward shift and the slope increased for the BD +placebo group relative to all other groups.

Conclusions: Consistent with our prior results, temporal perception was significantly different in the BD vs. HC groups. Poorer timing accuracy and precision in the BD+placebo group indicated that people with BD who do not use cannabis had altered temporal precision. Furthermore, THC administration exerted a unique effect on participants with BD, normalizing the clock speed of people with BD. These data are also consistent with prior research on the effects of acute THC administration on temporal perception. The small sample size of this study limits the power to detect interactive significant differences between groups, though the effect sizes were moderate. Thus, further investigation in a larger cohort is warranted. Future research should investigate whether cannabis treatment and/or its components can remediate temporal perception deficits in people with BD, potentially normalizing other behaviors

Keywords: cannabis, Temporal discrimination, cannabinoid
Disclosure: Nothing to disclose.

P110. ADDRESSING EFFICACY AND SAFETY OF MIXED AMPHETAMINE SALT AS AN ADJUNCTIVE THERAPY FOR BIPOLAR DEPRESSION

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Background: Depression is the prevailing pathological mood state of bipolar disorder and is associated with substantial morbidity and mortality. Despite high rates of inattention-poor focus in bipolar depression and ADHD comorbidity, there has been little systematic investigation of non-anti-manic mood stabilizers, such as stimulants, as a treatment intervention. MYDAYIS® (mixed d-amphetamine and l-amphetamine) is a central nervous system stimulant that is FDA approved and indicated for the treatment of attention deficit hyperactivity disorder (ADHD) in patients 13 years and older.

Methods: This placebo-controlled 2-site study was designed after the 2014 placebo controlled single site study of lisdexamfetamine in bipolar depression. 23 subjects were required per arm to achieve, using a two-sample t-test, significance level of 0.05 with 0.8 power. Given 2nd site and generous estimate of drop rates, we planned to enroll 90 subjects (45 subjects per site). Recruitment fell short of this target. Nonetheless, the achieved sample size ($n = 32$), yielded 80% power to detect a minimum effect size of $d \geq 1.03$.

Thirty-two bipolar (14 BPI, 18 BP-II) depressed patients (10M, 22F, mean age 38.2 years) who provided informed consent enrolled in an 8-week, randomized, double-blind, placebo-controlled study of adjunctive stimulant d-amphetamine/l-amphetamine (MYDAYIS®). The primary outcome measure was baseline to endpoint or at last observation carried forward of the Montgomery-Asberg Depression Rating Scale (MADRS). Secondary outcomes measures included: treatment response and remission (50% MADRS reduction, MADRS < 10), percentage of much or very much improved as measured by the Clinical Global Impression for Bipolar Disorder (CGI-BP), and baseline to endpoint or at last observation carried forward in the Quick Inventory for Depressive Symptomatology, clinician and self-rated (QIDS-C and QIDS-SR). Primary safety assessment was treatment-emergent manic symptoms as measured by the Young Mania Rating Scale (YMRS).

Results: Mixed d-amphetamine and l-amphetamine participants were slightly older than placebo participants (mean 42.7 vs 34.2; $p = 0.07$). Baseline symptoms (MADRS 27 ± 6.3 , QIDS-C 15.3 ± 3.2 , QIDS-SR 14.8 ± 4.7 , YMRS 4.5 ± 3.0) were consistent with moderate depression and absence of hypomania. There were no baseline demographic differences in sex, diagnosis subtype, all symptom rating scales, and concomitant medications (all $p \geq 0.13$). Participants receiving adjunctive stimulant d-amphetamine/l-amphetamine showed significantly greater improvement in depressive symptoms on the MADRS compared to placebo (median percent change: 65.4% vs. 31.8%; $p = 0.01$), with higher response (66.7% vs. 11.8%; $p = 0.003$) and remission rates (53.3% vs. 11.8%; $p = 0.02$). CGI-BP ratings indicated greater clinical improvement in the adjunctive stimulant d-amphetamine/l-amphetamine group (60.0% vs. 11.8%; $p = 0.008$). Clinician-rated QIDS-C scores improved significantly with adjunctive stimulant d-amphetamine/l-amphetamine (50.0% vs. 25.0%; $p = 0.01$), while self-reported QIDS-SR improvements did not reach significance

(40.7% vs. 21.7%; $p = 0.10$). No significant differences were observed in median percent change YMRS scores [stimulant d-amphetamine/l-amphetamine 63.6% vs. placebo 0.0%; $p = 0.14$; last YMRS score: 3.5(2.8) vs. 5.7(11.0)].

Conclusions: To our knowledge, this is the first study of a unique mixed amphetamine salt that suggests effective reduction in depression symptoms with no emergence of mania, hypomania, or psychosis. Studies are underway to identify clinical correlates of treatment response with a focus on anti-manic mood stabilizer, concurrent antidepressant therapy, presence or absence of atypical symptoms and inattention.

Keywords: Bipolar Disorder, stimulant, bipolar depression

Disclosure: Assurex Health, Grant, Self, Baszucki Group, Grant, Self, BD2 (Breakthrough Discoveries for Thriving with Bipolar Disorder), Grant, Self, Mayo Foundation, Grant, Self, Carnot Laboratories, Consultant, Self, American Physician Institute, Consultant, Self, Chymia LLC, Other Financial or Material Support, Self.

P111. NETWORK-LEVEL STRUCTURAL BRAIN ABNORMALITIES IN BIPOLAR DISORDER: A COORDINATE-BASED NETWORK MAPPING META-ANALYSIS

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Background: Bipolar disorder (BD) is a clinically and biologically heterogeneous illness, with onset across the lifespan and fluctuating mood states. Structural neuroimaging studies have consistently reported abnormalities in BD, yet findings remain variable and often conflicting. This inconsistency may reflect both the underlying clinical heterogeneity of BD and limitations of traditional meta-analytic approaches, which typically focus on isolated brain regions rather than distributed circuits. Circuit-level mapping may therefore provide greater clarity on convergent structural abnormalities in BD. To address this, we conducted a large-scale coordinate-based network mapping (CBNM) meta-analysis of whole-brain structural studies in BD.

Methods: We performed a systematic review and coordinate-based network meta-analysis in accordance with PRISMA criteria. Eligible studies included whole-brain T1-weighted voxel-based morphometry of grey matter volume and surface-based morphometry of cortical thickness reporting significant case-control differences. There was no age restriction for inclusion. Coordinates of structural alterations were extracted to generate study-specific seed maps. Using normative functional connectivity data from unrelated participants of the Human Connectome Project, dual regression was applied to identify networks connected to these seed regions. Study-level network maps were pooled to determine convergent patterns of network disruption, localized to Yeo functional networks (7-network parcellation) and major subcortical structures. Analyses were performed across the full sample and within subgroups defined by BD subtype (BD-I, BD-II), mood state (depressed, manic/hypomanic, euthymic), and above or below median age. Results are reported as Z scores reflecting the strength of cross-study convergence; values > 3 were considered statistically significant corresponding approximately to $p < 0.001$ (uncorrected), a threshold commonly applied in coordinate-based meta-analyses to denote robust cross-study convergence.

Results: We screened 11,356 records, reviewed 715 full texts, and included 131 case-control associations. Across the full sample, significant convergence ($Z > 3$) was observed in the visual

(L: $Z = 3.83$; R: $Z = 3.73$), dorsal attention (L: $Z = 3.20$; R: $Z = 3.07$), and control networks (L: $Z = 3.01$; R: $Z = 3.00$). In subgroup analyses, BD-I ($n = 77$) demonstrated significant convergence in visual, dorsal attention, and control networks, while BD-II ($n = 11$) significantly converged on visual and dorsal attention networks. In depression ($n = 23$), significant convergence extended across visual, dorsal and ventral attention, somatomotor, and control networks. Mania/hypomania ($n = 9$) had significant convergence in subcortical regions (hippocampus, putamen, caudate) and the default network, alongside visual and somatomotor systems. No significant effects were detected in euthymia ($n = 51$). Age-stratified analyses showed overlapping patterns: both younger (≤ 35 years, $n = 65$) and older (> 35 years, $n = 66$) groups demonstrated convergence in visual, dorsal attention, and control networks, with additional somatomotor involvement in younger samples.

Conclusions: This meta-analysis identifies consistent structural convergence in visual and attentional networks across BD, with additional network- and region-specific effects linked to clinical subtype and mood state. The absence of significant findings in euthymia highlights the dynamic, state-dependent nature of BD-related brain disruptions. Importantly, by situating abnormalities within distributed circuits rather than isolated regions, a network-based approach helps reconcile the heterogeneity observed across studies and highlights how diverse regional findings may reflect common underlying circuit dysfunction. These results support coordinate-based network mapping as a valuable framework for identifying circuit-level pathology in BD and guiding the development of stratified, network-informed interventions.

Keywords: Bipolar Disorder, meta-analysis, MRI

Disclosure: Nothing to disclose.

P112. ADVANCING DETECTION AND MEASUREMENT-BASED CARE OF MOOD PROBLEMS IN YOUTH: PAIRING NORMATIVE AND OUTPATIENT DATA

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Background: Measurement-based care (MBC) is increasingly adopted to guide diagnosis and treatment in child and adolescent psychiatry. Two key aspects are reliable detection of manic symptoms and accurate assessment of impact on functioning and quality of life (QoL). The Child Mania Rating Scale (CMRS-P) is widely used to evaluate manic symptoms, yet there are no nationally representative normative data. At the same time, the recently developed Nationwide Quality of Life Scale (NQLS) offers a brief parent-report tool for functional assessment, but its validity in clinical and population-based samples requires further study. This two-sample study combines psychometric evidence for the CMRS-P and NQLS to advance tools for pediatric mood disorder assessment.

Methods: Outpatient Cohort. Data came from 522 youth (39% female; M age = 10.6 years, SD = 3.5; range 5–18) enrolled from urban community and academic mental health clinics (NIH R01MH066647). Parents completed the full CMRS-P-21, from which the CMRS-P-10 was derived. Reliability (Cronbach's α , ω), precision, and correlations between scales and external validators were examined.

Normative Cohort. Nationally representative parent-report data were collected via survey panel for 1045 children aged 6–11 years (53% male). Parents completed the NQLS alongside measures of global impairment (N-Gauge), mania (CMRS-P10 and 21),

depression (VQIDS-5), suicidality (CHRT16), and internalizing symptoms (PSC-P-Int). Internal reliability and convergent validity were analyzed.

Results: Outpatient Cohort. CMRS-P-21 scores averaged 15.6 (SD = 10.6), with $\alpha = 0.90$ and $\omega = .92$. The CMRS-P-10 averaged 6.5 (SD = 5.0), with $\alpha = 0.79$ and $\omega = 0.79$. The two versions correlated strongly ($r = 0.96$, $p < 0.0001$). In terms of criterion validity, CMRS scores correlated $r = 0.39$ with bipolar diagnosis, .29 with any mood disorder, .34 with number of diagnoses, and -0.29 with global functioning. Convergent validity was high: $r = 0.66$ – 0.79 with other mania scales, versus 0.38 to 0.47 with externalizing, attention, or internalizing problems.

Normative Cohort. CMRS forms replicated excellent internal consistency and correlation. NQLS scores averaged 21.5 (SD = 4.5). Internal consistency was strong ($\alpha = 0.86$). CMRS-P-10 also correlated with external validators including depressive symptoms (VQIDS, $r = 0.56$), suicidality (CHRT, $r = 0.53$), emotional dysregulation (EDI, $r = 0.60$), ADHD symptoms (Vanderbilt, $r = 0.54$), internalizing symptoms (PSC-17, $r = .56$), and global impairment (N-Gauge, $r = 0.48$) (all $p < 0.001$). Modest but significant associations were observed with parent-reported diagnoses of depression, anxiety, ADHD, bipolar disorder, and disruptive behavior disorders ($r = .18$ – $.30$, $p < 0.001$). Higher CMRS-P scores were linked to greater service utilization, including medication and therapy, and a greater number of endorsed diagnoses ($r = 0.38$, $p < 0.001$). NQLS correlated negatively with manic (CMRS-P, $r = -.28$), depressive (VQIDS, $r = -0.40$), suicidal (CHRT, $r = -0.47$), and internalizing (PSC-P-Int, $r = -0.37$) symptoms (all $p < 0.0001$). Lower NQLS scores also correlated with diagnoses of depression and bipolar disorder, and with higher total disorder counts.

Conclusions: Across two large cohorts, findings demonstrate the reliability and validity of complementary MBC tools. The CMRS-P (full and brief) shows strong psychometric support in outpatient clinical populations, converging with symptoms, diagnoses, and service use. The NQLS provides a novel, psychometrically sound QoL assessment in a national community sample, capturing functional impairment linked to mood severity. Together, these instruments strengthen the assessment of mania severity and functional impact in youth. These are the first data using a nationally representative sample for both scales, and the pairing of an outpatient treatment-seeking sample with a normative sample greatly enhances the generalizability of results.

Future work should extend benchmarks for clinical change and diagnostic thresholds, and further integrate QoL assessment into MBC frameworks.

Keywords: Bipolar Disorder, measurement-based care, manic symptoms, quality of life

Disclosure: Joe Startup Technologies, Stock/Equity - Privately Held Company, Self, American Psychological Association, Royalties, Self, Guilford Press, Royalties, Self.

P113. AUTISTIC-LIKE TRAITS ARE ASSOCIATED WITH MOOD SYMPTOMS, NEUROCOGNITIVE FUNCTION, AND QUALITY OF LIFE IN PATIENTS WITH BIPOLAR DISORDER

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Background: Autistic-like traits are frequently elevated in individuals with bipolar disorder (BD), potentially reflecting shared neurodevelopmental mechanisms with autism spectrum disorder (ASD). Even in the absence of an ASD diagnosis, traits such as social communication difficulties and cognitive rigidity may

contribute to mood symptoms, cognitive impairment, and reduced quality of life (QOL). This study examined the associations among autistic-like traits, depressive symptoms, cognitive function, and QOL in individuals with BD.

Methods: Participants were 130 outpatients with BD (58 men, 72 women; median age = 34.1 years) at the National Center of Neurology and Psychiatry Hospital in Tokyo. Autistic-like traits were assessed with the Social Responsiveness Scale for Adults (SRS), depressive symptoms with the 17-item Hamilton Depression Rating Scale, cognition with the Brief Assessment of Cognition in Schizophrenia (BACS), and health-related QOL with the 36-Item Short-Form Health Survey (SF-36).

Results: Higher SRS scores were positively associated with depressive symptoms and negatively with cognitive performance, mental component summary (MCS), and role/social component summary (RCS) scores of the SF-36. Mediation analyses showed that autistic-like traits exerted a significant direct effect on MCS and indirect effects on MCS, RCS, and physical component summary (PCS) scores via depressive symptoms. Although autistic-like traits were related to poorer cognitive functioning, BACS scores did not directly predict QOL.

Conclusions: Autistic-like traits are associated with poorer mental health and indirectly contribute to diminished physical health and social functioning through depressive symptoms in BD. Assessing these traits may aid in developing personalized treatment strategies and improving functional outcomes.

Keywords: Quality of Life (QoL), psychological wellbeing, neurocognitive function, Depressive Mood

Disclosure: Boehringer Ingelheim, Advisory Board, Self, Takeda Pharmaceuticals, Consultant, Self.

P114. COGNITIVE–MOTIVATIONAL PROFILES IN YOUTH WITH AND WITHOUT FAMILIAL RISK FOR BIPOLAR DISORDER

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Background: Cognitive and motivational system (i.e., behavioral approach and inhibition systems) alterations are implicated in bipolar I disorder and may be detectable in individuals at familial risk. Identifying subgroups with distinct cognitive–motivational patterns could clarify heterogeneity in vulnerability.

Methods: Latent profile analysis (LPA) was conducted in 182 adolescents using scores from Cambridge Neuropsychological Test Automated Battery (CANTAB) tasks (memory, attention, executive function, psychomotor speed, emotion and social cognition) and Behavioral Inhibition/Activation System (BIS/BAS) scales. Multinomial logistic regression examined familial risk associations with profile membership, adjusting for demographics and childhood trauma (CTQ).

Results: A four-profile solution fit best: (1) Lower Cognitive Performance ($n = 17$); (2) Slow Reaction Time ($n = 23$); (3) Working Memory Deficits ($n = 47$); (4) Resilient ($n = 89$). Results showed that familial risk was associated with lower likelihood of being in Profiles 2 ($OR = 0.2$, $p = 0.05$) and 4 ($OR = 0.2$, $p = 0.03$) vs. Profile 1. Familial risk was also associated with lower likelihood of being in Profile 3 ($OR = 0.3$, $p = 0.07$) vs. 1 (marginally significant). When CTQ was added, these associations attenuated and became nonsignificant. Higher CTQ was independently associated with lower odds of Resilient membership ($OR = 0.5$, $p = 0.02$).

Conclusions: Familial risk was linked to a lower likelihood of resilient cognitive–motivational profiles, but this effect diminished after accounting for childhood trauma. Higher trauma was independently associated with reduced likelihood of resilience, suggesting early-life adversity may shape vulnerability profiles in bipolar disorder risk.

Keywords: latent profile analysis, behavioral approach system, behavioral inhibition system, cognition, bipolar disorder

Disclosure: Nothing to disclose.

P115. POLYGENIC RISK FOR BIPOLAR DISORDER IS ASSOCIATED WITH CORTICAL AND SUBCORTICAL STRUCTURAL DEFICITS IN ADHD YOUTH WITH FAMILIAL RISK FOR BIPOLAR I DISORDER

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Background: Bipolar I disorder (BD) is highly heritable, accounting for 60–85% of risk variance, and genome wide association studies (GWAS) indicate that BD liability is polygenic. Having a first-degree relative with BD is a robust risk factor for developing BD as well as other psychiatric disorders including attention-deficit/hyperactivity disorder (ADHD). Indeed, BD and ADHD have overlapping genetic liability, and the prevalence rate of ADHD in youth with a BD family history is significantly higher than youth without a BD family history. Longitudinal studies further indicate that the risk of developing BD among youth with ADHD is ten times higher compared to youth without ADHD. While these findings suggest that ADHD youth with a family history of BD are at greater risk for developing BD, the underlying neurobiological mechanisms remain poorly understood. Consistent with neurodevelopmental mechanisms, structural imaging studies have found that ADHD youth with a BD family history exhibit cortical and subcortical morphological deficits compared with ADHD youth without family BD history and healthy youth. However, the genetic contribution to these aberrant morphological changes have not been fully elucidated. BD polygenic risk scores (BD-PRS) are a weighted aggregate of multiple BD risk alleles derived from GWAS meta-analyses. Studies have identified higher BD-PRS in unaffected individuals with family history of BD, particularly those with early psychopathology. To extend these findings, the present study investigated associations between BD-PRS and cortical and subcortical structural metrics in psychostimulant-free ADHD youth with and without a first-degree relative with BD as well as a healthy comparison group.

Methods: ADHD youth (ages 10–18 years) with ('high-risk', HR) and without ('low-risk', LR) a first-degree relative with BD and typically developing healthy controls (HC) were enrolled. All ADHD youth met DSM-5 criteria for ADHD (any type), were stimulant-naïve or had no exposure to psychostimulants for at least 3 months prior to enrollment, and had no current DSM-5 mood, conduct, eating, or psychotic disorders. Subjects/parents were evaluated with Kiddie Schedule for Affective Disorders and Schizophrenia, and the Family Interview for Genetic Studies with the Structured Clinical Interview for DSM-5 on their first-degree relatives to assess for BD diagnosis. DNA was extracted from whole blood and genotyped using the Infinium Global Screening Array + PsychBooster. Following established protocols and standard quality control of genetic samples, polygenic risk scores for BD were constructed using PRS-CSx. High-resolution 3D T1-weighted images were acquired using a Philips 3.0 T MR scanner. The FreeSurfer image analysis suite was used to measure cortical/subcortical volume, thickness, and surface area. General linear models evaluated group differences in BD-PRS adjusting for age,

sex and the first four genetic principal components. Linear mixed models evaluated the relationship among MRI structural measures, group, and BD-PRS. Group differences were deemed significant with a threshold level of the false discovery rate (FDR) corrected $p < 0.05$.

Results: A total of 142 youth (mean age: 14.7 ± 2.6 , 37% female) were included in the analysis (HC $n = 36$, LR $n = 50$, HR, $n = 56$). There were no significant group differences in demographic variables. Differences between groups in BD-PRS were detected in the general linear model (main effect of group $p = 0.01$). Specifically, HR youth exhibited higher BD-PRS compared to LR youth ($p < 0.01$) and HC youth ($p = 0.02$), and BD-PRS did not differ between LR and HC youth ($p = 0.98$). Group differences were detected for total brain volume ($p = 0.02$), total cortical gray matter volume ($p < 0.001$), total subcortical gray matter volume ($p < 0.001$), total surface area ($p < 0.001$), and cortical thickness ($p = 0.01$). Compared to both LR and HC youth, HR youth had lower values for cortical gray matter volume ($p < 0.001$ and $p < 0.001$ respectively), total subcortical gray matter volume ($p < 0.001$ and $p < 0.001$, respectively), and total surface area ($p < 0.001$ and $p < 0.01$, respectively). HR youth also had lower values of mean cortical thickness compared to HC youth ($p = 0.04$), and smaller brain volumes compared to LR youth ($p = 0.03$). Among all subjects, BD-PRS was inversely correlated with bilateral orbitofrontal, middle frontal, insula, precuneus, caudate, and hippocampus volumes ($p < 0.05$). BD-PRS was also inversely correlated with left inferior frontal, right fusiform, right postcentral, right inferior parietal, left superior parietal, left supramarginal, right temporal (inferior, middle and superior), and left amygdala volumes ($p < 0.05$). For surface area, BD-PRS was inversely correlated left inferior frontal gyrus, bilateral orbitofrontal, left middle frontal, right anterior cingulate, right inferior parietal, right precuneus, left precuneus, left superior parietal, and left supramarginal areas. Regional cortical thickness metrics were not significantly associated with BD-PRS.

Conclusions: ADHD youth with familial risk for BD exhibit higher BD-PRS and cortical and subcortical structural deficits in regions implicated in the pathophysiology of BD. Associations between BD-PRS and these regional structural deficits support a polygenetic etiology, and future longitudinal studies are warranted to determine the impact of BD-PRS on neurofunctional as well as clinical trajectories.

Keywords: ADHD, Bipolar I disorder, Polygenetic Risk Score, Structural MRI

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P116. THETA NEURAL OSCILLATIONS AS A MARKER OF AFFECTIVE INHIBITORY CONTROL DEFICITS IN BIPOLAR DISORDER AND ALCOHOL USE

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Background: Individuals with bipolar disorder (BD) who have problematic alcohol use experience greater mood instability, higher suicide risk, and worse treatment outcomes. However, most

research on BD excludes people with problematic alcohol use, limiting progress in the identification and targeted treatment of shared neurobehavioral risk factors. This project aimed to identify EEG-based neurophysiological signatures of affective inhibitory control, a target neurobehavioral risk factor for BD and problematic drinking. We hypothesized that neural oscillations in theta, which underlie affective inhibitory control, would be altered in those with BD and would associate with drinking severity.

Methods: Individuals with BD I and II (BD, $n = 26$), subclinical BD (subBD, $n = 24$), and healthy controls ($n = 13$) underwent EEG recording during an Emotion Go/NoGo task. Time frequency decomposition was used to extract event-related theta-band (4–7 Hz) neural oscillatory power (Theta ERSP) over midline fronto-central areas during Go and NoGo trials. The Alcohol Use Disorder Identification Test (AUDIT) was administered as a continuous measure of drinking severity.

Results: Individuals with BD had lower theta ERSP during Go ($\beta = -1.14$, 95%CI $[-0.54-1.75]$, $p < 0.001$, $D = -1.28$) and NoGo trials ($\beta = -0.98$ $[-0.36$ to $-1.61]$, $p = 0.003$, $D = -0.92$) compared to HC. They did not differ from HC in the magnitude difference in theta ERSP between Go and NoGo trials. Individuals with subBD fell intermediate between HC and BD; however, they were not significantly altered compared to HC. Higher drinking severity was strongly associated with the magnitude difference in theta ERSP between Go and NoGo trials ($\beta = 0.42$ $[-0.01$ to $0.85]$, $p = 0.001$, $f^2 = 0.22$), but not associated with theta ERSP in Go and NoGo trials.

Conclusions: Results highlight an overall reduction in theta ERSP in those with BD and a specific deficit in those with higher drinking severity in which groups of neurons may fail to synchronize to effectively promote affective inhibitory control in NoGo trials. These results begin to suggest that individuals with BD who drink alcohol may particularly benefit from neuromodulation interventions that modulate theta-band activity. Future research should examine whether those with an alcohol use disorder (AUD) show the same generalized vs. specific alterations in theta-band oscillatory power.

Keywords: Bipolar Disorder, Alcohol, theta band oscillatory measures

Disclosure: Boehringer Ingelheim International GmbH, Consultant, Self.

P117. GABAPENTIN IMPROVES SLEEP, AS MEASURED BY ACTIGRAPHY AND SELF-REPORT, IN PEOPLE WITH CO-OCCURRING BIPOLAR AND ALCOHOL USE DISORDERS: RESULTS FROM A RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, CROSSOVER STUDY

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Background: It is estimated that nearly 40% of people with Bipolar Disorder (BD) develop Alcohol Use Disorder (AUD). Sleep and circadian rhythm disturbances are central to BD, and studies have shown that AUD is associated with sleep disturbances that can worsen BD symptoms. Previous evidence suggests that FDA-approved gabapentin and N-acetylcysteine (NAC) may be effective in managing some AUD symptoms (e.g., withdrawal, craving). Gabapentin has also been shown to improve primary insomnia in individuals struggling with sleep disorders. These pharmacological treatments are yet to be explored in the context of sleep outcomes in people with BD+AUD, however. We previously conducted an NIH/NIAAA-supported brief (1-week/condition),

randomized, double-blind, placebo-controlled, crossover study of adjunctive NAC (2400mg/day) and gabapentin (1200 mg/day). Here we present secondary sleep outcomes acquired via self-report and actigraphy, a non-invasive ambulatory technique for monitoring human rest/activity cycles. We hypothesized that gabapentin would improve sleep outcomes relative to placebo.

Methods: Fifty-four individuals with BD I or II and moderate to severe AUD were enrolled across a 5-year period, with daily use of mood-stabilizing medication a prerequisite for enrollment. Exclusions included serious medical illness, psychotic disorder, and medication dose changes $\geq 20\%$, ≤ 2 weeks before enrollment. Participants completed 3, 1-week conditions (i.e., gabapentin, NAC, placebo) in a randomized order. Each condition consisted of an in-person study visit including assessment of sleep quality (Pittsburgh Sleep Quality Index [PSQI], modified to past-week recall) and dispensing of medication (Day 1), titration to maximum dose (Days 1–5), and medication washout (Days 5–7). Throughout the 3-week study period, participants wore an Actiwatch (Philips Respironics) on their non-dominant wrist. Participants were instructed to press a button on their Actiwatch to generate event markers each time they went to sleep, woke up in the middle of the night and morning, took naps, and/or removed the device. Sleep outcomes, including Sleep Efficiency (SE), Sleep Onset Latency (SOL), Number of Awakenings (NWAK), Wake After Sleep Onset (WASO), and Total Sleep Time (TST), were calculated using Philips Actiware Software. Linear Mixed Models (LMM), each containing main effects of treatment condition and visit along with their interaction (i.e., to evaluate order effects), were conducted for each sleep variable in IBM SPSS software. Models involving PSQI additionally covaried for baseline PSQI scores.

Results: As reported previously, NAC and gabapentin were very well tolerated, medication adherence via urinary riboflavin detection was excellent, and the study blind was successfully maintained. Forty-eight BD + AUD participants with actigraphy data completed at least one experimental condition, providing a total of 124-weeks of valid actigraph data ($n = 39$ NAC, $n = 42$ Placebo, $n = 43$ Gabapentin). For SE ($n = 2$) and SOL ($n = 7$), additional condition outliers (i.e., $>Q3 + 3 \times IQR$ or $< Q1 - 3 \times IQR$) were removed prior to analysis to meet LMM assumptions. Treatment condition was significantly associated with SE ($F = 11.39$, $p < 0.001$), NWAK ($F = 7.28$, $p = 0.001$), and WASO ($F = 5.24$, $p = 0.008$), with pairwise comparisons demonstrating that only gabapentin was associated with higher SE ($\Delta[SE] = 3.67[0.96]$, $p < 0.001$, $SMD = 0.58$) and lower NWAK ($\Delta[SE] = -7.13[2.30]$, $p = 0.003$, $SMD = -0.55$) and WASO ($\Delta[SE] = -10.41[3.89]$, $p = 0.009$, -0.42) relative to placebo. In contrast, treatment condition was not associated with SOL ($F = 2.17$, $p = 0.122$) or TST ($F = 0.23$, $p = 0.792$). Concerning PSQI, one condition outlier was removed prior to analysis to meet LMM assumptions, and two participants missing baseline data were excluded from analyses. Controlling for baseline PSQI scores ($F = 4.87$, $p = 0.033$), treatment condition was significantly associated with posttreatment PSQI scores ($F = 3.68$, $p = 0.031$), with pairwise comparisons demonstrating that only gabapentin was associated with lower PSQI scores relative to placebo ($\Delta[SE] = -1.70[0.69]$, $p = 0.015$, $SMD = -0.47$). There was no evidence of condition order effects across analyses.

Conclusions: NAC and gabapentin were both safe and well-tolerated in BD + AUD participants, but only gabapentin significantly improved sleep outcomes as measured by actigraphy and self-report, whereas NAC did not separate from placebo on any measured sleep outcome. These initial findings suggest that gabapentin may represent a promising adjunctive medication for improving sleep maintenance among people with BD + AUD. Replication and extension of our findings across a longer dosing interval will be required to support this conclusion more definitively.

Keywords: Sleep disturbances, Bipolar Disorder, Dual Diagnosis, Alcohol Use Disorder - Treatment, Actigraphy

Disclosure: Nothing to disclose.

P118. COMORBID SUBSTANCE USE DISORDERS AMONG YOUTH WITH BIPOLAR DISORDER ARE ASSOCIATED WITH INCREASED CARDIOVASCULAR RISK

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Background: Substance use disorders (SUD) and cardiovascular disease (CVD) risk are both excessive and burdensome among adults and youth with bipolar disorder (BD). While the association between cardiovascular risk and SUD has been studied in adults, little is known about this topic in youth. The present study therefore examined the association of cardiovascular risk with SUD in youth with BD.

Methods: Participants included 212 youth with BD ($n = 72$ with SUD, $n = 140$ with no substance use) and healthy controls without SUD (HC; $n = 220$), ages 13–20 years. Multinomial logistic regression investigated the association of a composite cardiovascular risk score (comprised of blood pressure and body mass index [BMI]) with SUD, controlling for age and sex.

Results: Relative to youth with BD and no comorbid SUD, those with comorbid BD and SUD (OR = 1.30, 95% CI = [1.03, 1.65], $p = 0.026$) had higher cardiovascular risk scores, whereas HC had lower cardiovascular risk scores (OR = 0.73, 95% CI = [0.59, 0.90], $p = 0.004$). Each cardiovascular risk factor was examined independently. Relative to youth with BD and no comorbid SUD, those with comorbid BD and SUD had higher blood pressure (OR = 1.41, 95% CI = [1.02, 1.95], $p = 0.04$) whereas HC had lower blood pressure (OR = 0.72, 95% CI = [0.54, 0.96], $p = 0.03$) and lower BMI (OR = 0.63, 95% CI = [0.44, 0.92], $p = 0.02$).

Conclusions: Comorbid SUD among youth with BD is associated with higher cardiovascular risk. Future longitudinal studies are needed to examine the temporal association of these findings, in addition to the clinical factors and biological mechanisms that may account for this relationship.

Keywords: Bipolar Disorder, Youth, Substance Use Disorders, Blood Pressure, Body Mass Index

Disclosure: Nothing to disclose.

P119. MITOCHONDRIAL GENETIC VARIANTS ARE ASSOCIATED WITH CEREBRAL METABOLIC RATE OF OXYGEN BUT NOT CARDIORESPIRATORY FITNESS AMONG YOUTH WITH BIPOLAR DISORDER

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Background: Mitochondrial DNA (mtDNA) variants are associated with anomalous cerebral energy metabolism, and reduced cardiorespiratory fitness (CRF). Bipolar disorder (BD), which is associated with reduced CRF, anomalous cerebral blood flow and oxygen metabolism, is also associated with increased rates of mtDNA variants. We set out to examine mtDNA variants in relation to cerebral metabolic rate of oxygen consumption (CMRO2) and CRF in youth with BD.

Methods: Participants included 105 youth ($n = 55$ BD, $n = 50$ control group, CG), among which 97 had CMRO2 and 76 had CRF data. mtDNA common variants (i.e. minor allele frequency >5%) were derived from saliva. We generated an mtDNA variant functional impact (FI) score by performing functional analysis using Mutserve and summing the MutPred, Selection Score, and MitoTool scores. CRF was estimated during recumbent cycling. CMRO2 was calculated using Fick's principle, combining CBF with venous oxygen saturation (Yv) measured via T2-Relaxation-Under-Spin-Tagging (TRUST) MRI. We examined the main and interaction effect of FI score and individual mtDNA variants in relation to CRF and CMRO2, controlling for age and sex.

Results: Within BD, the presence of mt4216 variant was associated with lower CMRO2 ($F = 4.31$ $p = 0.04$), while the presence of mt14798 variant was associated with higher CMRO2 ($F = 4.12$ $p = 0.049$). There were no significant findings for CRF or among CG.

Conclusions: This study found that mtDNA variants were associated with CMRO2 among youth with BD, but not among CG. Present findings provide preliminary evidence for the role of mitochondrial genetics in cerebral oxygen metabolism in youth with BD.

Keywords: Bipolar Disorder, Mitochondrial DNA, youth, energy metabolism

Disclosure: Nothing to disclose.

P120. HIGHER POLYGENIC RISK FOR MITOCHONDRIAL DYSFUNCTION IS ASSOCIATED WITH LOWER CEREBRAL BLOOD FLOW IN YOUTH WITH BIPOLAR DISORDER

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Background: Bipolar disorder (BD) is a polygenic disease characterized by anomalous cerebral blood flow (CBF), mitochondrial dysfunction, and excessive cardiovascular risk. This study examined mitochondrial polygenic risk scores (Mito-PRS), derived from BD-PRS and cardiovascular PRS, in relation to CBF among youth with BD.

Methods: 100 participants (17.2 ± 1.6 years old; $n = 61$ BD, $n = 39$ controls) underwent arterial spin labelling magnetic resonance imaging to quantify CBF. Mito-PRSs were calculated using adult BD-PRS and myocardial infarction PRS genome-wide association summary statistics. Covariate-adjusted region-of-interest analyses examined the association of each Mito-PRS with global grey matter, anterior cingulate cortex, and amygdala CBF in the combined sample, and within diagnostic subgroups. Sex-stratified analyses and PRS-by-diagnosis interaction effects were also examined. Sensitivity analyses controlled for psychotropic medications and cardiovascular risk factors.

Results: In the overall sample, higher BD-derived Mito-PRS was associated with lower CBF in the global gray matter ($\beta = -0.29$, $p = 0.003$), anterior cingulate cortex ($\beta = -0.25$, $p = 0.01$) and amygdala ($\beta = -0.24$, $p = 0.01$). Similar findings were observed for the cardiovascular-derived Mito-PRS across all three regions-of-interest ($\beta[\text{range}] = -0.32$ to -0.24 , $p[\text{range}] = 0.001$ to 0.01). In the BD group, both Mito-PRSs were significantly associated with lower global CBF and marginally associated with lower amygdala CBF. The cardiovascular-derived Mito-PRS was also marginally associated with lower anterior cingulate cortex CBF in the BD group ($\beta = -0.26$, $p = 0.049$). Findings in the overall sample and the BD group were significant in females but not in males. Majority of findings remained significant when controlling for medications

and cardiovascular risk factors. There were no significant findings in the HC group, and no significant PRS-by-diagnosis interactions.

Conclusions: Higher polygenic risk for mitochondrial dysfunction, as implicated in BD and cardiovascular diseases, was associated with lower global and regional CBF in youth with BD, and particularly in females. Future longitudinal studies incorporating putative mediators of the observed findings are warranted.

Keywords: Bipolar Disorder, Cardiovascular Disease, Mitochondrial Dysfunction, Polygenetic Risk Score, Cerebral Blood Flow

Disclosure: Nothing to disclose.

P121. REPEATED KETAMINE INFUSIONS FOR TREATMENT RESISTANT BIPOLAR DISORDER: A RANDOMIZED, DOUBLE-BLIND, MIDAZOLAM-CONTROLLED PHASE II CLINICAL TRIAL (NCT05004896)

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Background: Treatment resistant bipolar depression (TRBD) is a significant clinical challenge with an urgent need for novel treatments. Growing evidence supports rapid and robust antidepressant effects with sub-anesthetic doses of intravenous (IV) ketamine for treatment resistant depression (TRD). The majority of completed randomized controlled trials (RCTs) to date have been in major depressive disorder (MDD) samples, excluding participants with history of mania, hypomania or psychosis. Only small, single dose pilot RCTs have evaluated ketamine for bipolar depression to establish proof-of-concept. No completed RCTs have evaluated the effects of a full acute course of IV ketamine for TRBD. As such, our primary objective was to evaluate the efficacy, safety and tolerability of an acute course (i.e., 4 infusions over 2 weeks) of IV ketamine for TRBD. Evaluating a full acute course (rather than only single infusion studies) is of critical importance to better understand the clinical utility of ketamine for TRBD. Additionally, while single dose studies showed minimal risk for treatment emergent mania or psychosis, it is possible that an acute course of ketamine holds greater risk for manic-switches compared to one single dose. As such, rigorous assessment of safety and tolerability is critical, with a special focus on risk of mania, hypomania and psychosis following an acute course of ketamine infusions.

Methods: We are conducting a multi-site, randomized, double-blind, midazolam-controlled, phase II clinical trial evaluating the efficacy, safety and tolerability of four flexibly dosed ketamine infusions (0.5–0.75 mg/kg infused over 40 min) for acute treatment of moderate to severe TRBD (including both BDI and BDII). Midazolam was used for the control arm to reduce risk of functional unblinding. Exclusion criteria include active psychosis, mania, substance use and severe medical comorbidity. The primary outcome is change in Montgomery-Asberg Depression Rating Scale (MADRS) scores from baseline to Day 14, using analysis of covariance (ANCOVA), with 14-day MADRS as the outcome and baseline MADRS and stratification variable (sex, bipolar type) as covariates. Secondary outcomes include response (>50% MADRS reduction) and remission (MADRS < 12) rates, safety (adverse events), tolerability (including treatment emergent mania), suicidality, anxiety, quality of life, function and duration of effects (to Day 28). Sex and gender-based analyses will also be conducted for all outcomes.

Of note, participants are assessed for adverse events daily during the acute course of treatment to carefully evaluate the

presence of any new symptoms, including manic symptoms or suicidality to thoroughly evaluate adverse events associated with ketamine, including transient effects that may have been missed with less frequent assessments.

Target enrolment is 70 participants ($n = 35$ per group randomized; assuming up to 10% drop out as seen previous studies) to be adequately powered for our primary outcome (baseline to Day 14 change in MADRS) to detect a minimum effect size of 0.6 (power = 0.8; $\alpha = 0.1$; standard for phase II trials), as seen in previous TRD ketamine studies. The power calculations and complete statistical analysis plan (SAP) was developed in close consultation with a team of senior clinical trial statisticians who will support all primary and secondary analyses for the trial.

This trial received local research ethics approval, a Health Canada No Objection Letter, Section 56 exemption for use of controlled substances and was registered with ClinicalTrials.gov: NCT05004896. This trial is funded by the Canadian Institute of Health Research (CIHR) with no industry/private involvement.

Results: Enrollment is currently ongoing with the trial to be completed by October 2025 (hard stop deadline due to funding) with 63 participants randomized to date. The mean age of the total sample is 45 (SD 11) with 53% female participants and mean baseline MADRS of 36 (standard deviation 5). To date, 26 participants have a diagnosis of bipolar I disorder and 37 are diagnosed with bipolar II disorder. No participants to date have experienced treatment emergent mania or psychosis. Efficacy (change in MADRS), safety, tolerability and key secondary outcome data (anxiety, suicidality, manic symptoms) will be presented at the conference as all data will be locked, cleaned and analyzed in November 2025, ensuring availability for presenting all outcome data at the January 2026 ANCP meeting.

Conclusions: There is an urgent need to evaluate the efficacy, safety and tolerability of an acute course of IV ketamine for TRBD. If results find ketamine to be efficacious and safe, this novel intervention will have the potential to address the current lack of sufficient pharmacotherapies for TRBD and hold hope for improving clinical outcomes in this difficult to treat population. As the first RCT evaluating an acute course of IV ketamine for TRBD, our trial will provide critical evidence to support or refute the use of IV ketamine for TRBD to inform clinical care guidelines and to determine if phase III testing is merited and feasible.

Keywords: (R,S)-ketamine, Bipolar Disorder, bipolar II disorder, Treatment-resistant depression, Psychedelics

Disclosure: Nothing to disclose.

P122. MULTIDIMENSIONAL NEURAL SIGNATURES OF MANIA AND DEPRESSION RISK: A REPLICATION AND EXTENSION ACROSS MOOD, COGNITION AND ENERGY DIMENSIONS IN THREE YOUNG ADULT SAMPLES

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Background: Robust reproducible neural markers of bipolar disorder (BD) risk are critical for early identification and improved differentiation from unipolar depression. The Mood Spectrum Self-Report–Lifetime version (MOODS-SR) has shown strong utility in parsing risk across BD and unipolar depression, particularly via its manic ‘mood’ domain. Our previous work found, across three independent young adult samples, replicated associations between approach emotion-related neural network response

patterns and lifetime risk for mania/hypomania and depression, as measured by the MOODS-SR manic and depressive mood domains: greater bilateral ventrolateral prefrontal cortex (vlPFC)–right(R) dorsolateral PFC (dlPFC) functional connectivity (FC) and reduced R caudate activity were associated with greater mania and depression risk, respectively, while greater bilateral amygdala–left(L) amygdala FC was associated with both greater mania and depression risk. It is unknown whether (a) these replicated neural patterns generalize across all MOODS-SR subdomains and (b) they represent domain-invariant and/or domain-specific markers of mood disorder risk. We aimed to determine whether these previously identified replicated neural patterns associated with the MOODS-SR ‘mood’ domains are specific to mood, or whether they also generalize to other dimensions of risk i.e., MOODS-SR manic and depressive ‘cognition’ and ‘energy’ subdomains, and finally whether these extensions also replicate across the three samples.

Methods: Three young adult samples (total $n = 299$) without BD completed a facial emotion processing fMRI task (discovery: male/female = 34/80, age = 21.60 ± 1.91 ; test sample 1: M/F = 30/83, age = 21.57 ± 2.09 ; test sample 2: M/F = 31/51, age = 23.43 ± 2.86). The previously replicated neural measures associated with the MOODS-SR mood domains (i.e., extracted parameter estimates of BOLD activity and FC to approach-related emotions from an anatomical region-of-interest mask supporting emotion processing/regulation ($pFWE < 0.05$, $k > 20$) were included as independent variables/IVs. Next, four parallel Poisson loglinear regression models identified the extent to which replicated neural IVs were significantly associated with each mania and depression risk domain, using MOODS-SR manic and depressive cognition and energy subdomain scores as model dependent variables. The IVs for the manic subdomain models were inter-amygdala FC and vlPFC–R dlPFC FC; the IVs for depressive subdomain models were inter-amygdala FC and R caudate activity. Results were FDR-corrected across models within samples.

Results: Manic Cognition: In the discovery sample, a Poisson loglinear regression model found that inter-amygdala FC and vlPFC–R dlPFC FC were positively associated with MOODS-SR manic cognition domain score ($qFDRs < 0.001$); both findings replicated in both test samples: inter-amygdala FC and vlPFC–R dlPFC FC were positively associated with manic cognition (all $qFDRs < 0.001$).

Manic Energy: In the discovery sample, a Poisson loglinear regression found that inter-amygdala FC ($qFDR < 0.001$) and vlPFC–R dlPFC FC ($qFDR = 0.002$) were positively associated with MOODS-SR manic energy domain score. The positive association between inter-amygdala FC and manic energy replicated in test sample 1 ($qFDR < .001$) only, while the positive association between vlPFC–R dlPFC FC and manic energy replicated in test sample 1 ($qFDR = 0.004$) and was marginally significant in test sample 2 before FDR ($P = .04/qFDR = 0.051$).

Depressive Cognition: In the discovery sample, a Poisson loglinear regression model found that R caudate activity was negatively, and inter-amygdala FC was positively, associated with MOODS-SR depressive cognition domain score ($qFDRs < 0.001$). Both findings replicated in both test samples: R caudate activity was negatively associated with depressive cognition (test sample 1 $qFDR < 0.001$; test sample 2 $qFDR = 0.011$) and inter-amygdala FC was positively associated with depressive cognition ($qFDRs < .001$).

Depressive Energy: In the discovery sample, a Poisson loglinear regression model found that R caudate activity was negatively, and inter-amygdala FC was positively, associated with MOODS-SR depressive energy domain score ($qFDRs < 0.001$). In both test samples, inter-amygdala FC was positively associated with depressive energy (test sample 1 $qFDR < 0.001$; test sample 2 $qFDR = 0.002$).

Conclusions: Our study replicates and extends prior work by demonstrating that specific patterns of neural activity and FC are differentially and consistently associated with lifetime mania/hypomania and depression risk across multiple symptom dimensions (mood, cognition, energy). Across analyses, inter-amygdala FC emerged as a robust, cross-dimensional marker most consistently and positively associated with mania and depression subdomains—suggesting a shared affective neural correlate of general mood disorder vulnerability. In contrast, vIPFC–R dIPFC FC was more consistently associated with manic cognition and energy, identifying it as a potential mania dimension-specific risk marker reflecting greater VAN/SN-CEN engagement, while reduced R caudate activity linked most consistently with depressive cognition, reflecting overall CEN disengagement. These results support a multidimensional model of mood disorder risk and underscore the utility of subdomain-level MOODS-SR analysis for identifying candidate neurobiological markers with potential predictive and diagnostic relevance.

Keywords: Bipolar Disorder risk, Depression risk, Biomarkers for Risk Assessment, fMRI Replication, Mania biomarker

Disclosure: Nothing to disclose.

P123. 2-ARACHIDONOYLGLYCEROL (2-AG) IN BIPOLAR DISORDER: A HUB FOR ARACHIDONIC ACID AND CALCIUM DYSREGULATION IN INDUCED PLURIPOTENT STEM CELL (IPSC)-DERIVED DOPAMINE NEURONS

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Background: The endocannabinoid (eCB) system is comprised of lipid signaling molecules, enzymatic machinery for biosynthesis/degradation, and their respective receptors. The most abundant eCB molecule in the brain, 2-arachidonoylglycerol (2-AG), is known to play a role in a number of physiological processes, including mood, hunger, sleep and more. eCB signaling also plays a role in neuroplasticity, and is a modulator of other key neurotransmitter pathways including through effects on calcium signaling. eCB ligands are known to be important regulators of dopaminergic neurotransmission, a key neuronal molecule involved in the pathophysiology of multiple psychiatric disorders. Targeting eCBs to improve psychiatric disorders remains a tantalizing prospect, and interest in the area has resurged with the development of eCB synthesis and degradation inhibitors that have been demonstrated to be safe in clinical trials. 2-AG degradation is also a source of arachidonic acid in the brain, suggesting a potential reservoir for important lipid mediators downstream of 2-AG.

The present study utilizes iPSCs from individuals with bipolar disorder (BD) and healthy controls to generate midbrain dopamine neurons following a small molecule differentiation protocol in an effort to understand the intersection of eCB biology and dopamine neuron function in BD. BD is a mood disorder which is characterized by drastic swings from states of low energy (depression) to high energy (mania/hypomania), with detrimental quality of life impacts for patients. Dopaminergic neurotransmission and calcium dysregulation are core tenets of BD pathophysiology, and eCB dysregulation in BD may provide a link between both signaling pathways.

Methods: Induced pluripotent stem cells from $n = 3$ individuals with BD and $n = 3$ healthy controls (all derived from female donors) were used to generate dopaminergic neurons following a small molecule protocol. The generated neurons were characterized by immunofluorescent staining for neuronal markers and multi-electrode array (MEA) to measure electrical firing. Gene expression of eCB-related genes was conducted using RT-qPCR.

An ELISA was used to measure 2-AG levels in cell culture media of neurons, and mass-spectrometry was used to measure 2-AG and anandamide (AEA) levels in dopamine neurons. Mass-spectrometry was further used to measure levels of arachidonic acid and a panel of related lipid metabolites in dopamine neurons. Inhibitors of 2-AG degradation and synthesis were used to investigate if 2-AG is a source for these lipid metabolites. Mitochondrial calcium buffering in response to 2-AG and 2-AG + rimonabant (a CB1 antagonist) was measured using mitochondrial calcium indicator dye Rhod-2AM. Unpaired two-tailed t test, multiple-comparison corrected t -tests, Mann-Whitney test, or one-way and two-way ANOVA were used to test for significance depending on the assay. Cohen's d score and Hedges g score were used to measure effect sizes.

Results: Immunofluorescent quantification of NURR1 and FOXA2 expression in dopamine neurons derived from $n = 3$ HC and BD patients revealed no significant differences in BD, while co-expression of TH, VGLUT2 and NEUN was significantly higher in BD neurons ($d = 1.00$, $p < 0.05$). MEA recording revealed no difference in mean firing rate in BD neurons, while ISI CV was lower in BD neurons ($g = 1.21$, $p < 0.05$). CB1 receptor mRNA was significantly decreased in BD dopaminergic neurons ($d = 1.22$, $p < 0.01$). ELISA results indicated an elevation of 2-AG levels in dopamine neuron media ($d = 1.055$, $p < 0.05$). Mass-spec detected no AEA in dopamine neurons, while 2-AG was elevated in BD ($g = 1.33$, $p < 0.01$). Multiple arachidonic acid-related lipids were upregulated in BD dopamine neurons, including arachidonic acid ($d = 2.49$, $p < 0.05$). Treatment with 100 nM MJN110 (2-AG degradation inhibitor) for 8 hours did not reduce arachidonic acid levels in BD neurons. Treatment with 500 nM DO34 (2-AG synthesis inhibitor) for 8 h reduced arachidonic acid levels ($d = 1.18$, $p < 0.05$). Mitochondrial calcium was not affected by 200 nM 2-AG in dopamine neurons, while pre-treatment with 1 μ M rimonabant followed by 2-AG increased mitochondrial calcium in HC neurons ($g = 0.51$, $p < 0.001$). Rimonabant treatment alone reduced average calcium peak amplitude in BD neurons ($g = 0.85$, $p < 0.05$).

Conclusions: Dopamine neurons have been efficiently generated from HC and BD iPSCs, although differences in co-expression of VGLUT2 and TH may indicate differences in maturation rate in BD. The co-expression of VGLUT2 and TH suggests these neurons are similar to those found in the VTA. 2-AG levels in BD dopamine neurons are disrupted, and results from ELISA and mass-spec indicate that both total 2-AG and released 2-AG are higher in BD. Arachidonic acid and its related metabolites are upregulated in BD, and some success was observed in decreasing arachidonic acid levels by inhibiting 2-AG synthesis. Furthermore, mitochondrial calcium in response to CB1 antagonism is dysregulated in BD neurons, with rimonabant treatment decreasing calcium transient amplitude. This may be as a result of changes to CB1 receptor expression levels in BD neurons. Research is ongoing to determine the mechanisms underlying elevated 2-AG levels.

Keywords: iPSC-derived neurons, Bipolar Disorder, Endocannabinoids

Disclosure: Nothing to disclose.

P124. SINGLE-CELL MULTI-COHORT TRANSCRIPTOMIC DISSECTION OF BIPOLAR DISORDER

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Background: Bipolar disorder (BD) is a highly prevalent psychiatric disease, affecting 1–2% of the global population, and

individuals with BD experience significant mortality due to suicide. Despite this burden, the pathophysiology of BD remains incompletely understood, with diagnosis based on a heterogeneous set of clinical symptoms, requiring mania with or without additional mood or psychotic symptoms. Therapeutic options remain limited, with effective treatment of depression in BD being especially difficult. Advancing our understanding of BD's neurobiological mechanisms is essential for identifying new therapeutic targets in this highly heritable and polygenic disorder.

Methods: This single nucleus RNA sequencing study measured genome-wide gene expression in nuclei isolated from human prefrontal cortex of 44 postmortem donors with and 44 donors without BD across two independently assayed cohorts including both male and female donors. Differential gene expression analysis of these data integrated results from multiple complementary statistical approaches (MAST-RE, Nebula, edgeR). Downstream analyses included identification of impacted biological pathways (fgsea), integration with GWAS data (MAGMA), transcription factor binding motif analysis within coexpressed gene modules (hdWGCNA), cell-cell communication analysis (CellChat), and identification of potentially causal genes using Mendelian Randomization (TwoSampleMR).

Results: We identified 1932 unique differentially expressed genes across 4524 gene-cell type pairs, with the greatest degree of dysregulation in deep layer excitatory neurons. Dysregulated gene sets impact neurodevelopment and synapse relevant pathways, as well as intracellular signaling including inositol signaling pathways. Implicated transcription factors were identified and validated using CUT and Run, and intercellular communication analysis found overall increased strength of signaling through a decreased number of ligand-receptor pairs. Mendelian Randomization analysis nominated 17 differentially expressed genes as potentially causal of BD pathology, implicating the mitogen activated protein kinase and calcium calmodulin pathways in the disease process.

Conclusions: The single-nucleus transcriptomic analysis presented here provides necessary context for existing bulk tissue data and genetic knowledge, advancing our understanding of BD. The identification of relevant genes and pathways involved in BD pathogenesis paves the way for future research on novel therapeutic targets and facilitates mechanistic studies.

Keywords: Bipolar Disorder, Single nucleus RNA sequencing, Postmortem Human Brain Tissue, Mendelian Randomization

Disclosure: Nothing to disclose.

P125. DESIGNING THE BIPOLAR DISORDER ACTION NETWORK: A LEARNING HEALTH NETWORK

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Background: The system of care in the United States for people with bipolar disorder is fragmented and disjointed. The gaps are that methods of assessment are inconsistent, treatments vary widely and are frequently inconsistent with the best available guidelines, and at a population level, not only have outcomes not improved over the past 20 years, but care is not coordinated, outcomes are not tracked, and patients have no input in the design of their care. Healthcare systems work in silos and miss the opportunity to learn together to advance a common agenda to improve outcomes.

Methods: To address these gaps, we designed a Learning Health Network (LHN) for bipolar disorder. LHNs use a network-based organizational structure to facilitate collaborative, data-driven improvements in population health outcomes. They promote a culture of continuous learning, data sharing, and innovation, with a focus on achieving outcomes that matter to all stakeholders and accelerating the rate of improvement.

We used a structured, idealized system design process, considering the multiple organizations and their constituencies as a whole system and all participants in the system to identify gaps and potential solutions. The Bipolar Disorder LHN design process had four objectives: (1) Build a community by fostering a shared commitment to improving health and well-being among stakeholders, (2) Identify and test changes in care organization to enhance system performance, (3) Develop leaders and cultivate their skills in systems thinking and quality improvement (QI), and (4) Develop a rapid learning system by establishing measures to track the processes and outcomes of BP care, and the structures and actions to continuously evaluate and optimize the performance of the system.

Results: We engaged 17 healthcare systems and a design team of approximately 50 individuals representing patients, caregivers, clinicians, researchers, and advocacy organizations. We also offered quality improvement (QI) training to a subset of 9 of the prospective clinical site leaders. Design activities focused on four levels of the system: the patient and family, the clinical care organization, the community, and the network of health systems. Three design meetings were held to foster collaboration and generate ideas. We developed the network's vision, mission, and performance measures at all levels. An environmental scan identified existing evidence and innovations, and we adopted a patient journey map to align potential system changes with the patient experience. We generated design concepts and rated them based on potential impact and feasibility. We documented a theory of change using Key Driver Diagrams and logic models, developed core governance policies and processes, as well as other infrastructure, including data capture tools and a registry. A "blueprint" of system changes was developed to illustrate the connections between drivers of better clinical care and the patient journey. We organized measures in a dashboard to provide network participants with a "common operating picture", enabling situational awareness and collaborative decision-making. We also trained participants in co-production and developed an evaluation plan and a sustainability framework. Overall, we built an organization with a culture of humility, generosity, curiosity, and innovation.

Conclusions: Using a deliberate design process, we established an LHN organization (including collaborative relationships, design, structures, theories, tools, systems of work, and measures) to change the system of care to get better outcomes for people living with bipolar disorder.

Keywords: Bipolar Disorder, Learning Health Network, Healthcare System Change

Disclosure: Nothing to disclose.

P126. UPS TO DOWNS: PREDICTORS AND OUTCOMES OF POST-MANIC/MIXED EPISODE DEPRESSION ASSESSED IN ELECTRONIC HEALTH RECORDS

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Background: Depressive symptoms within weeks to months of a pure manic or mixed episode are common, and are associated

with relapse, hospitalisation and delayed recovery. However, "Post-Manic/mixed Episode Depression" (PMMD) remains an under-recognised syndrome.

Post-manic/mixed episode depression (PMMD) is under-recognised and largely overlooked by major clinical guidelines. However, depressive symptoms following mania are associated with relapse, hospitalisation and delayed recovery. Further, PMMD coincides with major suicide factors; bipolar depression (during which 1 in 3 attempt suicide), and post-discharge from acute care (3-times risk within 1 year than any other time).

PMMD is biologically interesting. A mood episode sequence of mania-depression-interval of euthymia (MDI sequence) is the strongest factor associated with lithium response ($OR = 4.3$, 95% CI: 2.61–6.97). PMMD is the first part of this sequence.

Previous work indicates that 30–40% of people with bipolar experience an MDI sequence (which proxies PMMD). However, this may be an underestimate, as MDI is usually only studied as a secondary outcome. There have essentially been no biological studies.

The aims of the present study were to use data from electronic health records to define the prevalence and clinical correlates of PMMD, and identify potential predictors.

Methods: We define a PMMD episode as the emergence of depression within 6 months of a mixed/manic diagnosis, and consider those who have experienced PMMD at some point in their history as a "PMMD group". As a comparator group ("noPMMD"), we consider people who have had both manic/mixed and depressive episodes, with the latter always > 6 months after a manic/mixed episode.

We examined the Neuroblu dataset of mental healthcare electronic health records, including 35 million patients across the United States. We identified $n = 11,261$ had both manic/mixed and depressive episodes (total episodes = 50,639), of which $n = 6540$ met criteria for the PMMD group, and $n = 4721$ for noPMMD.

We first compared the PMMD and noPMMD groups on the severity of depression, number of mood episodes, number of healthcare contacts, and comorbidity.

We also performed a within-group PMMD analysis to investigate factors during or before manic/mixed episodes that may dynamically increase risk of PMMD. We focused on hospital-associated episodes due to this data being higher quality. We included demographic and hospital visit-related covariates.

Both sexes were included in the study.

Results: PMMD was common: 46% of people who experienced a manic episode and 60% of people who experienced a mixed episode developed PMMD.

Comparing the PMMD and noPMMD groups, the PMMD group had twice as many mood episodes as the noPMMD group (PMMD: mean = 7.0, SD = 7.0; noPMMD: mean = 3.4, SD = 2.7, $p < 1e-16$) and 1.2-times the number of healthcare contacts (PMMD total visits: mean = 211.0, SD = 274.0; noPMMD total visits: 171.0, SD = 250.0, $p = 3.9e-11$). The PMMD group tended to be more unwell during mania/mixed episodes (mean CGI-S increase of 0.50, $p = 1.9e-4$) and depressive episodes (mean CGI-S increase of 0.52, $p = 7.2e-12$). The PMMD group was also more likely to have any ICD-10 major code (average $OR = 1.5$; SD(mean logOR) = 0.2; binomial test $p < 1e-16$) – an index of comorbidity. The PMMD group was also more likely to meet criteria for rapid cycling (Fisher test $p < 1e-16$) and MDI sequence (Fisher test $p = 1.1e-9$).

Within the PMMD group, lithium during a manic episode was associated with lower risk of PMMD in the following 180 days ($OR = 0.50$, SE = 0.29, $p = 0.016$) and was associated with a longer period until the next depressive episode (competing risks $HR = 0.74$, SE = 0.14, $p = 0.026$). Antiepileptic mood stabilisers (e.g., valproate, lamotrigine, carbamazepine) were protective against depression after manic ($OR = 0.55$, SE = 0.22, $p = 0.006$)

and mixed episodes ($OR = 0.68$, SE = 0.16, $p = 0.020$), but only modified time to depression after mixed episodes ($HR = 0.77$, SE = 0.07, $p = 5.4e-5$). For mixed episodes, a depressive episode in the previous 180 days was associated with subsequent PMMD.

Conclusions: PMMD is common and is associated with increased mental and physical comorbidity and healthcare contacts. Lithium during a manic episode may prevent subsequent PMMD, whereas other antiepileptic mood stabilisers may prevent PMMD after a mixed episode.

Keywords: Bipolar Disorder, electronic health record (EHR), Bipolar I Depression, Recovery

Disclosure: Nothing to disclose.

P127. THE ROLE OF THE MESOLIMBIC DOPAMINE SYSTEM IN BIPOLAR DISORDER

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Background: Bipolar disorder (BD) is characterized by recurrent episodes of mania and depression that are exacerbated by stress, yet the neurobiological mechanisms underlying stress-induced fluctuations in arousal remain poorly understood. The mesolimbic pathway, specifically dopamine (DA) projections from the ventral tegmental area (VTA) to the nucleus accumbens (NAc), is a critical overlapping node in emotion and arousal circuitries, and its dysregulation has been implicated in BD. The HYPER rat, which spontaneously arose in a colony of outbred Sprague-Dawley animals and has been selectively bred for extreme psychomotor hyperactivity (manic-like) and hypoactivity (depression-like) in response to stressors over 80+ generations, serves as a unique model for investigating the underlying mechanisms within the DA VTA-NAc circuit that may be involved in psychiatric disorders such as BD characterized by disturbances in stress-induced regulation of arousal.

Methods: We assessed mesolimbic DA function in male and female HYPER and wild-type (WT) Sprague-Dawley rats using amphetamine-induced locomotor activity and c-fos immunohistochemistry. RNAscope in situ hybridization targeting DA-related genes was performed in the VTA and NAc during baseline and stress-induced hyperactivity periods in HYPER and time-matched WT rats. Bulk RNA sequencing of the VTA and whole genome sequencing were used to define transcriptional and genomic differences between HYPER and WT rats.

Results: HYPER rats displayed a blunted locomotor response to amphetamine but elevated c-fos expression in the NAc, consistent with sensitized DA receptor activity. RNAscope revealed a trend toward lower expression of D2 and D3 receptors in the NAc of HYPER rats compared to WT. Whole genome sequencing identified two amino acid-altering variants in the dopamine receptor D3 (*Drd3*) gene unique to HYPER rats. To test their functional impact, we transfected HEK293 cells with WT or mutant *Drd3* constructs and measured receptor-mediated cAMP signaling using a GloSensor assay. Dose-response analyses revealed mutation-specific alterations in *Drd3* function. Ongoing analyses are examining transcriptional signatures in the VTA and activity-dependent gene expression within mesolimbic nodes.

Conclusions: Results from these experiments will enhance our understanding of how alterations in mesolimbic DA transmission, including reduced DA receptor expression, altered signaling dynamics, and *Drd3* variants, contribute to stress-induced psychiatric phenotypes relevant to BD.

Keywords: Bipolar Disorder, Dopamine, Arousal

Disclosure: Nothing to disclose.

P128. SYNAPTIC PROTEOMES IN MOUSE AND HUMAN BRAIN*Mazdak Bradberry, Rajesh Soni, David Sulzer, Maura Dupont**Columbia University, New York, New York, United States*

Background: Schizophrenia is a common and often devastating brain illness marked by psychosis, withdrawal, and cognitive impairment. Genetic and epidemiological studies suggest that many distinct risk factors and biological pathways ultimately lead to the development of schizophrenia symptoms. The 'synaptic hypothesis' of schizophrenia suggests that dysfunction at synapses represents a primary pathological process, or point of convergence, in the brain pathology of schizophrenia. Many studies have suggested changes in synaptic composition or function in schizophrenia, but clear and consistent neuropathological correlates of this illness remain poorly established. Proteomic analysis represents an opportunity to define changes in synaptic composition in rich molecular detail, but previous work defining synaptic proteome changes in schizophrenia from postmortem brain tissue has been limited by relatively low specificity for synaptic proteins.

Methods: We demonstrate the adaptation of proximity-labeling proteomics analysis to brain synapses. In formaldehyde-fixed slices from mouse or human brain, presynaptic boutons were biotinylated using a horseradish peroxidase-SV2 monoclonal antibody conjugate (HRP-SV2mAb), and postsynaptic densities were biotinylated using a horseradish peroxidase-PSD95 nanobody conjugate (HRP-PSDnb). For proteomics analysis, brain slices were then lysed, and the dissolved biotinylated proteins were isolated using streptavidin magnetic beads. Isolated proteins were digested with trypsin and analyzed by liquid chromatography and mass spectrometry. Slices incubated without HRP conjugates were used as negative controls to account for endogenously biotinylated proteins and other sources of background. For biotin localization analysis, slices were stained using fluorescent streptavidin and imaged by confocal microscopy. Cell types of origin for selected proteins were predicted using data from single-cell RNA-sequencing databases hosted by the Allen Institute for Brain Science.

Results: Synaptic proteomes obtained by proximity labeling yielded >1000 proteins specifically enriched in biotinylated samples. Proteins corresponding to a range of biological functions including glycolysis, aerobic respiration, redox homeostasis, membrane trafficking, and neurotransmission were quantified. The abundances of detected proteins correlated well with previous studies of the synaptic proteome using alternative preparation methods. Proteins predicted to originate from a variety of cell types including neurons and astrocytes were observed. Confocal microscopy demonstrated the expected synaptic localization for biotin deposited by either HRP-SV2mAb or HRP-PSDnb, confirming that the detected proteins primarily originate from synaptic material.

Conclusions: Synaptic proteomics by proximity labeling represents a straightforward approach toward the deep molecular characterization of synaptic composition. It is anticipated that these techniques will be applied to postmortem brain tissue from subjects with schizophrenia, along with animal models of schizophrenia risk, to clarify the synaptic pathophysiology associated with this brain illness.

Keywords: Schizophrenia and related disorders (SRD), spatial proteomics, neuroinflammation, Postmortem Human Brain Tissue

Disclosure: Nothing to disclose.

P129. CELLULAR RESOLUTION SPATIAL AND TRANSCRIPTOMIC CHARACTERIZATION OF THE HUMAN LATERAL SEPTUM ACROSS NEUROANATOMICAL AXES*Robert Phillips, Madeline Abramson, Yufeng Du, Svitlana Bach, Sarah Maguire, Ryan Miller, Ishbel Del Rosario, McKenna Romac, Joel**Kleinman, Thomas Hyde, Stephanie Hicks, Vincent Costa, Keri Martinowich, Stephanie Cereceo Page**Lieber Institute for Brain Development, Johns Hopkins University School of Medicine, Baltimore, Maryland, United States*

Background: The lateral septum (LS) is a GABAergic basal forebrain region that exhibits functional control over social behavior. Within the rodent brain, the LS is subdivided into dorsal (LSd), intermediate (LSi), and ventral (LSv) domains that exhibit unique transcriptional signatures and connectivity patterns. Transcriptionally distinct cell types within these domains are spatially distributed along the anterior-posterior, dorsoventral, and mediolateral axes and control specific aspects of social behavior. While the transcriptional heterogeneity and spatial topography of rodent LS cell types have been characterized, our understanding of the distribution of LS cell types in the human brain is limited due to freezing-induced alterations in neuroanatomical presentation and limited resolution of existing atlases.

Methods: To address these challenges, we set out to map the spatial topography and transcriptional heterogeneity of molecularly distinct cell types within the human lateral septum using in situ sequencing with a custom 300 gene panel. To develop the custom gene panel, we used marker genes from previously published mouse and human single nucleus RNA-sequencing (snRNA-seq) data. We also generated snRNA-seq data from spatially resolved tissue punches of the rhesus macaque LS, collecting over 37,000 cells and allowing us to retain information regarding the cell types present within LSi, LSd, and LSv domains. Following the design of the 300 custom gene panel, we generated the first detailed spatial and transcriptional atlas of the human LS. Across 2 donors (1M/1F), we collected tissue sections containing LS every 500 microns spanning the entire extent of the anterior-posterior axis, while retaining mediolateral and dorsoventral orientation. In addition to in situ spatial transcriptomics, we also generated single nucleus RNA-sequencing (snRNA-seq) data from the same donors at specific points along the anterior-posterior axis, providing the ability to generate whole transcriptome data while retaining anterior-posterior orientation. Using snRNA-seq data from rhesus-macaque and mouse, we correlated t-statistics for every gene within each cluster with the t-statistics in the spatial transcriptomics data to understand the conservation of molecular signatures of LS cell types across species.

Results: Unsupervised, spatially-aware clustering with BANKSY identified several transcriptionally and spatially distinct spatial domains representing the LSi, LSd, and LSv subdivisions of the human LS. We find that OXTR, the gene encoding the Oxytocin receptor is selectively expressed within LSv, while PRKCD, the gene encoding Protein Kinase C Delta, and CHODL, the gene encoding Chondrolectin, is selectively expressed within LSd and LSi, respectively. The ability to retain anterior-posterior orientation across sections allowed us to identify that both genes and transcriptionally distinct LS cell types change across the anterior-posterior axis. We find that genes such as FREM2 are more highly expressed within posterior sections, while NKX2-1 is more highly expressed within middle sections. In addition to the identification of LS subdomains, we also identify transcriptionally distinct domains representing the dorsal striatum, bed nucleus of stria terminalis (BNST), and medial septum (MS). Correlation of t-statistics identifies several conserved transcriptional signatures including those cells that exhibit expression of TRPC4 and DGKG.

Conclusions: These experiments provide the first high resolution spatial atlas of the human LS, advancing our knowledge of its cytoarchitecture and neuronal heterogeneity, as well as that of neighboring regions. Cross-species analyses identify both conserved and divergent cell types and marker genes. Together these

data provide a robust resource to further understand how molecularly distinct cell types are involved in social behaviors, and how dysregulation of these cell types may be involved in the development of psychiatric diseases.

Keywords: Lateral Septum, spatial transcriptomics, human and non-human primates, Single nucleus RNA sequencing

Disclosure: Nothing to disclose.

P130. CORTICAL GABAERGIC NEURON DYSREGULATION IN SCHIZOPHRENIA IS AGE DEPENDENT

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Background: Cortical GABAergic neuron dysregulation is implicated in schizophrenia (SCZ), but it remains unclear whether these changes are due to altered cell proportions or per-cell changes in mRNA expression.

Methods: We analyzed 14 bulk and cell type-specific RNAseq datasets from 1,408 individuals (672 SCZ cases, 736 controls) across three neocortical regions. We deconvolved GABAergic cell subtype proportions from bulk RNA-seq and benchmarked them against single-nucleus RNAseq and stereological densities from matched donors. We assessed SCZ- and age-associated changes in cell proportions and per-cell gene expression.

Results: SCZ was associated with altered proportions of neocortical parvalbumin (PVALB) and somatostatin (SST) cells, dependent on the subject's age at death. Younger SCZ cases (age < 70 years) showed reduced PVALB and SST cell proportions, while older cases showed unchanged or increased proportions compared to controls. Earlier onset SCZ, associated with more severe clinical symptoms, was linked to greater reductions in these cell types. Additionally, there was robust evidence for reduced per-cell SST and VIP mRNA among younger cases with SCZ.

Conclusions: These findings suggest that SCZ is associated with complex, age-dependent alterations in GABAergic neurons, particularly affecting PVALB and SST cells. Our study underscores the importance of age-stratified analyses in SCZ, suggesting that distinct pathological processes underlie GABAergic neuron dysregulation across different age- and symptom-severity groups and warranting tailored therapeutic approaches.

Keywords: Molecular neuropathology, Single-cell RNA sequencing, Somatostatin-expressing interneurons, Parvalbumin neurons, meta-analysis

Disclosure: Nothing to disclose.

P131. HUMAN HIPPOCAMPUS CELL GENOMIC PROFILES IN NEUROTYPICAL SUBJECTS AND MAJOR DEPRESSION

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Background: In the rodent hippocampus dentate gyrus (DG), adult neurogenesis produces new granule cells (GC) by activating

quiescent neural progenitors (QNP or Type I) that start dividing (ANP or Type II), differentiate into intermediate progenitors or neuroblasts (INP or Type III), and start maturing into immature GC (imGC) and then mature GC.

Human adult neurogenesis, first demonstrated in the DG subgranular zone (SGZ) and along the ventricles' subventricular zone (SVZ) of 57–72 years old cancer patients, remains controversial, even in studies implementing single nuclei RNA sequencing (snRNA-seq).

Single nucleus multiome sequencing (snMultiome-seq) co-profiling gene expression and chromatin accessibility has not been employed in human hippocampus, and allows better classification of cells types and resolving developmental trajectories given that locus accessibility precedes gene expression during cell differentiation.

Impaired adult neurogenesis and hippocampus plasticity may contribute to the pathogenesis of Major Depressive Disorder (MDD). People with MDD have fewer neural progenitors and GC, and those treated with antidepressants have more ANPs and GCs than untreated subjects. In rodents, adult neurogenesis supports stress recovery and memory, enhances hippocampus plasticity and rewiring, and prevents depressive-like behaviors.

We aimed to determine if a neurogenic niche exists in human hippocampus and what are the cell-type-specific or subfield-specific differentially expressed genes (DEG), differentially accessible chromatin regions (DAR), and differentially expressed proteins (DEPs) in MDD hippocampus.

Methods: For identifying rare cell types, we analyzed the largest number of nuclei (495,037) using the highest sequencing depth than previously done. To determine the anatomical location of each cell type in the hippocampus subfields, we performed spatial transcriptomics using Visium and Xenium.

To determine if the identified DEGs translate into DEPs, we performed shotgun proteomics in a separate sample. All assays and analyses were performed on anterior hippocampus proper (N = 36 CTRL age 14–74, 7 females; N = 26 MDD age 17–76, 8 females). Inclusion criteria: clear toxicology, except for benzodiazepines, no psychotropic drugs prescription for at least three months prior to death, sudden death without prolonged agonal state, and high-quality brain tissue based on RNA integrity number, tissue pH, and neuropathological assessment.

Results: We identified 31 unsupervised snMultiome cell clusters based on canonical gene markers and integrating our data with published human hippocampus snRNA-seq databases.

On Visium and Xenium slides, the snMultiome GC cell cluster mapped onto the GCL, the various excitatory neuron (ExN) clusters mapped on Cornu Ammonis (CA) and subiculum, inhibitory neurons (InN) were found in SGZ and hilus, astrocytes (Astro) in the strata oriens and lacunosum-moleculare, oligodendrocytes (Oligo) in the stratum radiatum that contains axons, ependyma (Epe) and choroid plexus (CP) correctly mapped on SVZ and choroid plexus areas, and endothelial (Endo), vascular leptomeningeal (VLMC), microglia (Micro), and T cells (TC) were found spread across the hippocampus subfields.

The neurogenic cell trajectory was identified by sub clustering GC and Astro, and using Monocle cell trajectory inference, which showed a trajectory between Astro and GC subclusters, going from Astro subtypes that express QNP markers, to cells that express mitotic markers (ANP), to GC subclusters expressing INP markers, followed by imGC and finally GCs.

Cell-type-specific gene expression and chromatin accessibility are altered in MDD.

Across all cell types, we identified 1248 DEGs in MDD compared with CTRL. InN.PENK.MME and GC.1 contributed most DEGs, with 566 (45.35%) and 379 (30.37%) respectively. ExN.CA1-2.FIBCD1.FNDC1 had 147 DEGs (11.78%), ExN.proximalSUB.-MUC19.GLP2R had 47 (3.77%), Astro.1 had 23 (1.84%), Oligo.1 had 13 (1.04%), Oligo.2 and OPC both had 12 (0.96%).

ExN.SUB.IQCF3.KRT17 and GC.2 had 10 (0.80%), ependyma had 8 (0.64%), InN.SST.NDNF and InN.SST.NPY both had 5 (0.40%), micro had 4 (0.32%), and GC.3 had 2 (0.16%). ExN.distalSUB.MSLN-LANKRD34B, ExN.SUB.SMYD1.THEMIS, ExN.CA3-4.GUCA1C.MYPN, GC.4, and VLMC had one DEG each (0.08%). Clusters ExN.SUB.AB-CA12.QRFPR, InN.PVALB.LHX1, InN.LHX6.NOS1, InN.VIP.CALB2, GC.5, TC, Endo, CP, Astro.2, and Oligo.3 had no DEGs.

In immature cell subclusters, there were 948 MDD DEGs.

Regarding DARs, Astro.1 had the most, with 78 hits (24.22%), following by Oligo.1 with 61 (18.94%), GC.1 with 55 (17.08%), ExN.CA1-2.FIBCD1.FNDC1 with 48 (14.91%), and Oligo.1 with 25 (7.76%). ExN.SUB.IQCF3.KRT17 (2.80%) had 9 DARs and Endo contributed 7 (2.17%). ExN.CA3-4.GUCA1C.MYPN and GC.2 had 5 each (1.55%), and OPC had 4 (1.24%). All other clusters contributed <1% DARs each to the total: Astro.2, CP, InN.PENK.MME, and Micro had 3 each. GC.3, InN.LHX6.NOS1, InN.SST.NPY, and TC has 2 each. Ependyma, ExN.distalSUB.MSLN-LANKRD34B, InN.PVALB.LHX1, and InN.SST.NDNF had one DAR.

Some hippocampus DEPs were altered in the same direction of cluster-specific DEGs in MDD vs. controls.

Conclusions: This multiome profiling of human hippocampus supports the presence of a neurogenic niche. Gene ontology analyses suggest altered molecular pathways in MDD involving cellular development, metabolism, signaling, apoptosis, and inflammation. Targets leads are being tested for drugs and biomarker discovery.

Keywords: Hippocampal neurogenesis, Major Depressive Disorder (MDD), RNA-seq, Proteomics, ATAC-seq

Disclosure: Nothing to disclose.

P132. STUDYING THE RELATIONSHIP BETWEEN PLACENTA AND RISK FOR SCHIZOPHRENIA USING IPSC-DERIVED TROPHOBLAST CULTURES

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Background: We previously found that genomic risk for schizophrenia (SZ) interacts with early life complications in affecting trajectories of risk for the disorder [PMIDs: 29808008, 33558239]. Further, we found that the placental genes associated with SZ risk are linked with the nutrient-sensing capabilities and invasiveness of the trophoblast (TB) [PMID: 37188697]. Here, we leverage an in vitro iPSC-derived model to study alterations of placental development and functions in patients affected with SZ with high genomic risk compared with neurotypical controls (NTC) with low genomic risk.

Methods: 23 fibroblasts from patients affected with SZ (N = 13) and NTC (N = 10) were reprogrammed in iPSCs. TB stem cells (TSC) were obtained from iPSCs after naïve-like resetting and induction of trophoctoderm lineage. TSCs were differentiated in syncytiotrophoblasts (STB) and extravillous trophoblasts (EVT). TB qualification was confirmed by RNAseq and marker expression. We performed functional analyses, studying proliferation, nutrient sensing, and metabolic activity of TSCs, the fusion index and hCG secretion of STBs. To detect differentially expressed genes (DEGs), we used linear mixed models, adjusting for RIN, mapping rate, sex, and cell line. DEGs were defined based on a false discovery rate (FDR) < 0.05. Statistical comparisons in functional assays were performed using a t-test.

Results: RNAseq data and marker expression validated the identity of TSCs, STB and EVT. We detected 92 differentially

expressed genes in TSCs, 89 in STBs, 13 in EVTs. These genes were consistently enriched for pathways associated with metabolic processes, highlighting, in SZ, an alteration in membrane and energy dynamics, also related to mitochondria and respiratory chain reaction. TSCs from patients with SZ show decreased level of glucose uptake; the impairment was greater in TSCs from male patients, and even greater under hypoxia ($p < 0.001$). The fusion index was higher in STBs from patients with SZ, but only in females ($p < 0.05$). Consistently, STBs from female patients secrete more hCG ($p < 0.05$). We are currently investigating mitochondrial metabolic activity of TB cultures from patients with SZ and NTCs and the impact of their extracellular vesicles on neural growth and differentiation.

Conclusions: Human iPSC-derived models of TB development and function are promising in the investigation of how the placenta might contribute to SZ. The identification of placental mechanisms of risk for SZ can facilitate the development of strategies of prenatal prevention of SZ, aimed at preserving the contribution of placenta to brain development.

Keywords: Schizophrenia (SZ), placenta stem cell models, Genomics

Disclosure: Nothing to disclose.

P133. SETD1A ORCHESTRATES PSYCHIATRIC GENE NETWORKS INVOLVED IN GENOMIC STABILITY AND SYNAPTIC FUNCTION IN BOTH RARE AND SPORADIC SCHIZOPHRENIA

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Background: Genome-wide association studies (GWAS) have identified 287 common variant loci associated with schizophrenia (SCZ) risk. While individual variants confer modest effects, their cumulative impact is substantial. The histone H3K4 methylation pathway represents a prominent biological pathway enriched for SCZ-associated variants. Genes encoding H3K4 methyltransferases, demethylases, and readers are frequently implicated in neurodevelopmental disorders (NDDs), suggesting a critical role for H3K4me3-associated pathways in human brain development. Beyond common variants, rare copy number variants and loss-of-function (LoF) mutations in individual genes also contribute significantly to SCZ risk. Rare heterozygous LoF mutations in SETD1A represent among the strongest single-gene risk factors for SCZ and are similarly implicated in NDDs. SETD1A encodes a methyltransferase that catalyzes H3K4me2/3 within the COMPASS complex. A recurrent patient mutation, SETD1A c.4582-2delAG, disrupts the exon 16 splice acceptor and has been identified across multiple SCZ and NDD cohorts. Despite compelling genetic evidence, SETD1A's molecular targets and function in the developing human brain remain poorly characterized. Whether SETD1A regulatory networks converge on SCZ polygenic risk loci or contribute to sporadic SCZ pathogenesis in the broader population remains unknown. Here, we investigate the consequences of SETD1A haploinsufficiency during early human neurodevelopment and its role in SCZ pathogenesis.

Methods: We introduced heterozygous LoF mutations (SETD1A +/-) into two neurotypical male iPSC lines using CRISPR/Cas9-mediated genome editing. We generated isogenic SETD1A +/- lines, including two lines carrying the recurrent

patient-specific c.4582-2delAG mutation at the exon 16 splice acceptor site, which causes intron retention and a premature stop codon in exon 16 (SETD1A c.4582-2delAG/+). Isogenic iPSC lines were differentiated into 2D neural progenitor cells (NPCs) and neurons, as well as 3D cortical organoids, to explore morphological and functional alterations caused by SETD1A haploinsufficiency. We performed bulk and single-cell transcriptome analyses on these neuronal models. To understand the mechanism of SETD1A-mediated gene expression regulation, we identified genome-wide SETD1A binding sites by CUT and Tag analysis in 2D NPCs, neurons, and postmortem brain tissue from three male fetal cortices in the second trimester. To investigate whether altered SETD1A activity might contribute to SCZ pathogenesis beyond rare LoF mutations, we evaluated the expression of SETD1A and its downstream targets in adult individuals with SCZ who do not carry SETD1A LoF variants, using large-scale postmortem cortical transcriptomic datasets from PsychENCODE and BrainSeq.

Results: We demonstrated that SETD1A preferentially binds to promoters of polygenic risk loci for psychiatric disorders, particularly those regulating chromatin remodeling, DNA repair, and synaptic function. SETD1A also binds to DNA damage-prone sites in both NPCs and postmitotic neurons. SETD1A haploinsufficiency caused aberrant cell cycle progression and accelerated neurogenesis in NPCs, as well as reduced neuronal complexity and DNA damage accumulation in postmitotic neurons. These defects were rescued by inhibiting the H3K4me2/3 demethylase KDM5. In postmortem SCZ cortical tissue, individuals lacking SETD1A LoF mutations exhibited reduced SETD1A expression compared to neurotypical controls, associated with downregulation of SETD1A-regulated SCZ GWAS genes. These findings implicate dysfunction of the SETD1A-H3K4me pathway in sporadic SCZ cases.

Conclusions: We integrated isogenic neuronal models carrying a patient-specific SETD1A c.4582-2delAG mutation, epigenomic profiling of human prenatal brain and neuronal cells, and statistical genetic analyses to dissect SETD1A's role in SCZ pathogenesis. Our findings demonstrate that SETD1A directly regulates SCZ GWAS-linked genes involved in synaptic signaling and DNA repair, providing a convergent molecular mechanism across rare and common SCZ risk. Our analyses also demonstrate that reduced SETD1A activity contributes to SCZ pathogenesis in a broader subset of patients. Even in the absence of rare LoF variants, lower SETD1A expression, which may result from common genetic variation or environmental factors, leads to downregulation of SETD1A target genes at SCZ risk loci. These genes are enriched for synaptic and genomic stability functions, suggesting that disruption of SETD1A-dependent regulation may represent a convergent mechanism in SCZ pathogenesis. Overall, the present study supports a model in which SETD1A haploinsufficiency causes transcriptional dysregulation and developmental genomic instability that propagates through neural lineages, leading to progressive dysfunction over time. These findings may bridge developmental and aging-related mechanisms underlying SCZ and highlight SETD1A as a key regulator of brain health across the lifespan.

Keywords: Induced pluripotent stem cells (iPSCs), Brain organoids, Rare genetic variants, Schizophrenia (SCZ)

Disclosure: Nothing to disclose.

P134. HUMAN IPSC-DERIVED OLIGODENDROCYTES IMPLICATE MATURATION DEFICITS IN NEUROPSYCHIATRIC DISEASE

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Background: Myelination is a continuous process within the central nervous system (CNS) that shapes neuronal activity to coordinate relevant circuitry. Deficits in this process resulting in hypomyelination or demyelination have been linked to cognitive deficits, neurodegenerative disorders, and multiple neuropsychiatric disorders including major depressive disorder, bipolar disorder, and schizophrenia. In addition, activity-dependent myelination of dopaminergic axons emanating from the ventral tegmental area has recently been shown to promote opioid reward learning in mice, demonstrating bidirectional modulation of myelination is relevant to disease. Therefore, unraveling the molecular mechanisms that shape myelin in the CNS may help identify novel therapeutic targets to treat neuropsychiatric disease.

TCF4 is a basic helix-loop-helix transcription factor that regulates the development of multiple neural cell subtypes. In mouse oligodendrocyte precursor cells (OPCs), TCF4 heterodimerizes with OLIG2 and promotes their differentiation into mature, myelinating oligodendrocytes (OLs) in a cell autonomous manner. TCF4 knockout (KO) mouse studies show additional circuit level alterations such as decreased myelin quality within the CNS, alterations to signal propagation across the corpus callosum, and increased locomotor activity in a novel open field. Mutations in TCF4 are also implicated in human disease; heterozygous loss-of-function mutations are causative for the neurodevelopmental disorder, Pitt-Hopkins Syndrome, and rare missense mutations have been associated with intellectual disability, autism spectrum disorders, and schizophrenia. Thus, modulating TCF4 expression may provide a molecular handle to investigate the cellular mechanisms underlying OL biology. Here, we set out to KO TCF4 and assess its role on human OL development and myelination using iPSC-derived OLs.

Methods: Human iPSCs were differentiated into OLs using transient SOX10 overexpression following established protocols and validated using qRT-PCR and immunocytochemistry (ICC). TCF4 KO iPSC lines were generated by inserting a premature stop codon into exon 5 of the long TCF-B isoform using CRISPR/Cas9 gene-editing. Gene-edited hiPSCs were subsequently differentiated into OLs and TCF4 expression was measured using qRT-PCR and western blot analysis. The maturation of TCF4-edited OLs was assessed using ICC. Data were analyzed using a one-way ANOVA with a Tukey correction for multiple comparisons and graphed as the mean $\pm$ SD. The significance level was set at $p < 0.05$. RNA sequencing was conducted on OLs isolated using anti-O4 microbeads. Differential gene expression analysis was performed using DESeq2 with a significance threshold set at $\text{padj} < 0.05$.

Results: Validation of the SOX10 differentiation demonstrated transient upregulation of genes associated with OPCs (PDGFRA, TCF4, and OLIG2) that was replaced with the later expression of mature OL genes (CSPG4, PLP1, MBP, MOG, and CLND11) as measured with qRT-PCR analysis ($n = 4$ differentiations). The temporal increase in MBP RNA expression was coordinated with an increase in protein expression as measured by ICC ($n = 3$, 1 differentiation). Furthermore, these OLs attached and extended processes when cultured on polymer nanofibers.

TCF4 KO hiPSCs were differentiated into OLs to validate the effective knockdown of TCF4. RNA expression of long TCF4 isoforms was significantly decreased in heterozygous and homozygous KO OLs, but total levels of TCF4 were unaltered ($n = 9$ –12 from 3 differentiations, 3–4 lines/genotype). Western blot analysis revealed the overall protein expression of TCF4 was decreased in heterozygous and homozygous KO lines due to the absence of multiple isoforms ($n = 12$ –16 from 4 differentiations, 3–4 lines/genotype).

The maturation of TCF4 KO OLs was assessed by ICC. The percentage of cells that expressed the pan-OL genes OLIG2 and O4 were not different between genotypes at multiple timepoints. On the other hand, the percentage of cells that express MBP was

significantly decreased in homozygous KO lines compared to WT ($n=24\text{--}32$ from 4 differentiations, 3–4 lines/genotype). RNA expression of multiple known mature OL genes was additionally decreased in KO OLs, but only MOG was among the 42 significant differentially expressed genes following correction for multiple comparisons ($n=9\text{--}12$ from 3 differentiations, 3–4 lines/genotype).

Conclusions: We have developed a human iPSC-based platform to assess the development and maturation of human OLs. Human iPSC-derived OLs robustly express known OL markers and extend processes along polymer nanofibers, indicating they hold the capacity to myelinate in vitro. Gene-edited TCF4 KO OLs showed a deficit in maturation reproducing those observations from KO mouse studies and a recent publication that used an alternative human iPSC differentiation approach. Importantly, this maturation deficit was not explained by a decreased commitment of differentiating cells to the OL lineage. Our findings confirm OL dysfunction is relevant for neuropsychiatric disease and demonstrate these iPSC-derived OLs will serve as a useful model to investigate cellular mechanisms underlying OL biology. Future experiments will establish a co-culture system with human iPSC-derived neurons and develop in vitro readouts of myelination necessary to explore how disease-related alterations to OL development shape neuronal function and activity.

Keywords: Induced pluripotent stem cells (iPSCs), oligodendrocytes, TCF4

Disclosure: Johnson and Johnson, Employee, Self.

P135. SINGLE-NUCLEUS RNA SEQUENCING OF CHOLINERGIC NEURONS IN THE HUMAN BASAL FOREBRAIN

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Background: The cerebral cortex and other forebrain regions receive their cholinergic input from a small population of cholinergic projection neurons in the basal forebrain. Dysfunction in the brain's cholinergic system has been implicated in psychiatric and neurological disorders such as schizophrenia and Alzheimer's disease, but few studies have characterized human brain cholinergic neurons at the molecular level.

Methods: We have been developing a method to isolate cholinergic neurons from the postmortem human forebrain for single-nucleus transcriptomics. Postmortem human basal forebrain tissue was obtained from neurotypical brain donors. Nuclei of cholinergic neurons were isolated from brain tissue samples using antibody staining and fluorescence-activated nuclear sorting. Single-nucleus RNA sequencing was performed using standard droplet-based protocols.

Results: Preliminary results indicate that cholinergic nuclei are rare in the human basal forebrain but they can be enriched by fluorescence-activated nuclear sorting. Single-nucleus RNA sequencing revealed two molecularly distinct clusters of cholinergic neurons in the human basal forebrain which differ in the expression of thousands of genes. Preliminary immunostaining results show that these two subtypes of cholinergic neurons are spatially intermingled in the human basal forebrain.

Conclusions: Preliminary data indicate that postmortem human forebrain cholinergic neurons can be isolated for single-nucleus RNA sequencing and that the human basal forebrain contains two molecular subtypes of cholinergic neurons. Ongoing work will aim to characterize the function of these two cell types and examine

their molecular alterations in brain donors with schizophrenia and other neuropsychiatric disorders.

Keywords: Single nucleus RNA sequencing, Cholinergic system, Postmortem Brain Tissue

Disclosure: Nothing to disclose.

P136. CROSS-DISORDER DYSREGULATION OF INTERCELLULAR COMMUNICATION REVEALS CONSERVED MECHANISMS IN HUMAN BRAIN DISORDERS

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Background: Cell-cell interactions (CCI) regulate brain homeostasis and mediate transcriptomic dysregulation in both neurodegenerative diseases (NDDs) and neuropsychiatric disorders (NPDs). Despite distinct clinical phenotypes, converging evidence suggests that NDDs and NPDs share molecular and cellular mechanisms, particularly at the level of intercellular signaling. Although prior studies have analyzed CCI in brain disorders, cross-disorder comparisons remain limited. To address this, we leveraged PsychAD, a population-scale single-nucleus RNA-seq resource developed by our group comprising over 6 million nuclei from the dorsolateral prefrontal cortex (DLPFC) of 1,494 human donors. We integrated statistical modeling with spatial validation to construct one of the largest and most robust cross-disorder atlases of dysregulated CCI in the human brain.

Methods: We applied the LIANA framework to infer CCIs from PsychAD across eight NDDs and NPDs, including Alzheimer's disease and related dementias (AD/ADRD), schizophrenia (SCZ), and bipolar disorder (BD). The cohort covers postnatal ages between 0 and 108 years, roughly equal numbers of males ($n=723$) and females ($n=771$), covers a diverse genetic background and over 30% of the donors ($n=509$) were of non-European ancestry. Differential analysis was performed using linear mixed-effects modeling, which accounted for both biological and technical covariates. We benchmarked across technical replicates and tissue sources, characterized shared versus disorder-specific effects, and validated the spatial proximity of prioritized CCIs with Xenium transcriptomics on ($n=11$) DLPFC samples. Functional interpretation included Gene Ontology enrichment and GWAS concordance tests.

Results: We inferred 38.9 million CCIs across 27 neuronal, glial, and immune cell types. After QC, 33,687 unique CCIs were tested across 8 disorders and 1494 donors. Of these, 14% of significantly differentially expressed ($FDR < 0.05$, $|\log_2FC| > 0.5$) CCI were shared across two or more disorders, highlighting conserved cross-disorder mechanisms. NDDs exhibited a predominant upregulation of microglia-vascular CCIs, whereas NPDs showed dysregulation of neuronal-astrocyte and neuronal-vascular CCIs. Shared effects largely featured downregulated inhibitory neuron subclasses (SST+ and LAMP5 RELN+). Specificity analysis confirmed consistent patterns of cross-disorder dysregulation. Pathway enrichment identified neuronal adhesion and synaptic maintenance as conserved dysregulated processes, with disorder-specific effects in microglia: lipid metabolism pathways in AD versus transport pathways in SCZ/BD. GWAS integration revealed significant enrichment of receptor-side CCI genes for disease risk loci, particularly for AD and SCZ GWAS. Spatial validation showed

that high-confidence CCI, as predicted by gene expression, exhibited colocalization with shorter mean intercellular distances than low-confidence CCIs.

Conclusions: This work defines a robust cross-disorder atlas of dysregulated CCI in the human brain, revealing both shared neuronal processes and disorder-specific microglial programs enriched for genetic risk. Our results demonstrate that intercellular communication is a critical component of disease convergence across psychiatric and neurodegenerative conditions. By integrating large-scale single-nucleus RNA-seq with statistical modeling, GWAS concordance, and spatial transcriptomics, this study provides a reproducible framework for identifying therapeutic targets and biomarkers at the systems-level of intercellular communication.

Keywords: Interactomes, Neurodegenerative Disease, Neuropsychiatric Disorders, CROSS DISORDERS, single-nucleus RNA-sequencing

Disclosure: Nothing to disclose.

P137. H3 SEROTONYLATION REGULATES DEVELOPMENTAL GENE EXPRESSION IN THE MPFC CONTRIBUTING TO BEHAVIORAL RESPONSE TO EARLY LIFE ADVERSITY

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Background: The serotonergic (5HTergic) system and its homeostasis in early life are critical to establishing the proper architecture of the developing brain. Early life stress (ELS) can alter these precise developmental trajectories and increase the lifetime risk for depression. Studies from our lab have established a novel ELS paradigm using a combination of maternal separation with limited nesting, that leads to a priming effect on stress susceptibility following subsequent exposures to stress in adulthood. Historically, the actions of 5HT were thought to be mediated exclusively through ligand-receptor interactions. However, our lab recently established a novel role for 5HT in the epigenetic regulation of gene expression. Specifically, 5HT forms covalent bonds with histone H3 tail where it stabilizes the adjacent H3 lysine 4 trimethylation post-translational modification (PTM). This combinatorial mark, termed H3 serotonylation (H3 Ser.) is critical for permissive gene expression in the neonatal developing brain both in vitro and in vivo. H3 Ser. has been functionally linked to responses to stress in adulthood. However, functional roles for H3 Ser. during postnatal neurodevelopment and impacts of aberrant environmental stimuli during early life have largely been unexplored.

Methods: We employed immunosorbent assays to examine 5HT levels across postnatal neurodevelopment in the medial prefrontal cortex (mPFC) – a region receiving dense 5HTergic innervation – with and without ELS. After examining differences in regional and developmental abundance of 5HT, we sought to investigate changes in H3 Ser. genomic localization across normal and perturbed neurodevelopment. To do this we leveraged fluorescent activated nuclei sorting (FANS)-using antibodies against the neuron-specific antigen, NeuN, to isolate neuronal- vs. glial-enriched mPFC cell populations in both male and female mice at each of our developmental time points of interest. I coupled this approach with C and R (Cleavage Under Targets and Release Using Nuclease)-sequencing for H3 Ser. on both cell populations from mPFC to directly assess epigenomic H3 Ser. enrichment patterns during normal postnatal development and following perturbation by ELS (3–4 biological replicates/group (three animals pooled/replicate). To first decipher the developmental trajectories of H3 Ser.

in both control and ELS animals, I performed differential enrichment analysis on FANS mPFC tissues from all time points assessed, anchored to the earliest development time point in the study (P10), using DiffBind (significance cut off: FDR < 0.1 and FC > |1.5|). Bioinformatic analysis was used to further dissect these results. Finally, immunohistochemistry was used to determine the cell-type specific roles of H3 Ser. in cell-type differentiation.

Results: We found mPFC 5HT levels fluctuate during postnatal neurodevelopment of mice and ELS decreases levels of 5HT acutely in adolescence ($p = 0.0188$), an effect not seen in adulthood ($p = 0.9761$; $n = 3-4/\text{group}$). FANS-coupled C and R revealed that normal developmental differences – as well as those perturbed by ELS – were more pronounced in males vs. females in both neurons and glia. Strikingly, in glia, ELS increased the number of differential loci by 150-fold in adolescence with 7927 differential enriched sites, representing the most pronounced differential enrichment observed across all conditions. These differential binding sites were largely gained in comparison to juvenile time points (P10) and were found to be distinct from age-matched controls ($p > 0.99$). These differentially enriched loci are enriched for genes involved in the regulation of chromatin remodeling, transcription, and post-translational protein modifications and are largely located at promoter regions. Cell-type deconvolution analysis revealed, that of the 7927 differential sites induced by ELS in NeuN- fractions at P21, 26.2% occurred within cell type-specific loci, with microglia, astrocytes, mature oligodendrocytes, oligodendrocyte progenitor cells, and endothelial cells all represented. Notably, upstream transcription factor analyses of ELS-induced differential peaks in glia at adolescence (P21) predicted OLIG2, (oligodendrocyte transcription factor 2) – a master regulator for oligodendrocyte lineage specification, which regulates key stages of early OL development – as a top associated transcription factor. Interestingly, in males, H3 Ser. displays increased Olig2 promoter binding in response to ELS at adolescence (P21) compared to age-matched controls. Immunohistochemistry (IHC) also revealed that Olig2+ cells show a trend toward increased H3 Ser. intensity ($n = 2/\text{group}$, $p = 0.04$) and the number of Olig2+ cells ($p = 0.06$). We also find an increased number of oligodendrocyte precursor cells with no change in the number of mature oligodendrocytes ($p = 0.04$ and 0.6444 respectively; $n = 4/\text{group}$). This is the first evidence to examine H3 Ser.'s role in oligodendrogenesis.

Conclusions: These findings provide novel insight into how H3 serotonylation regulates neurodevelopment and the mechanisms by which disruptions to this PTM cause aberrant pathophysiological states. We demonstrate normal; neurodevelopmental patterns of H3 serotonylation enrichment are reorganized by early life stress in a cell type-specific manner. This data provides novel evidence for the roles of H3 serotonylation in the regulation of oligodendrogenesis demonstrating novel roles for serotonin.

Keywords: Early life stress (ELS), Epigenetics, oligodendrocytes

Disclosure: Nothing to disclose.

P138. BEYOND D2: EXPLORING THE FUNCTION OF NUCLEUS ACCUMBENS DOPAMINE D3 RECEPTORS IN ANTIPSYCHOTIC ACTION

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Background: Second generation antipsychotics (SGAs) are widely used treatments for people with serious mental illnesses and other neuropsychiatric conditions. Research into antipsychotic mechanisms of action have predominantly focused on their actions at D2 dopamine receptors (D2Rs) given our more involved understanding of these G-protein coupled receptors (GPCRs). Many of these SGAs also bind D3 dopamine receptors (D3R), a subset of

the Gi-coupled D2R family, and some binding with higher affinity compared to D2Rs. Given recent advances in molecular biology and pharmacology, we now have the tools to dissect out the function of D3R independent of D2R using selective agonists/antagonists and genetic deletion in mice. A convergent area of interest lies in the nucleus accumbens where D3Rs are enriched in contrast to areas in the dorsal striatum where D2Rs are predominant. Our understanding of the role of the nucleus accumbens in reward learning and motivation makes it an attractive target relevant for the treatment of many neuropsychiatric disorders.

Methods: For slice electrophysiology, whole-cell current- and voltage-clamp recordings are obtained from nucleus accumbens medium spiny neurons in acute coronal slices prepared from p25–75 mice. D3R-expressing neurons will be targeted by crossing D3-Cre mice with Ai14 reporter mice, which carry a floxed td-Tomato fluorophore that can be visualized with 880 nm 2-photon excitation. These cells will be compared to Cre lines that report expression of D1 and D2. To assess D3R function, we measure intrinsic excitability properties, miniEPSCs/IPSCs, and evoked EPSCs/IPSCs in conditions with wash-on of D3R selective agonists/antagonists (PD128907, BAK4–54) and D3R-binding SGAs (cariprazine, quetiapine, clozapine). We also utilize constitutive D3-KO mice as well as conditional D3-cKO mice after stereotaxic injection into the nucleus accumbens with AAV-Cre-GFP. For imaging experiments, isoforms of the D1, D2 and D3 receptors are spatially and quantitatively assessed with RNA-in situ hybridization (HCR).

Results: Data from ex vivo slice electrophysiological experiments analyzing spontaneous miniature events in D3R-expressing medium spiny neurons (MSNs) of the NAc reveal a decrease in miniature IPSC (mIPSC) amplitude following the addition of a selective D3R agonist (PD128907) ($p < 0.05$) in a paired t-test. No significant change was observed in miniature EPSC (mEPSC) amplitude or frequency after D3R agonist addition. These findings suggest that D3R activation in the NAc modulates inhibitory activity in MSNs, likely through downstream regulation of GABAA receptors. In evoked experiments, a similar reduction in IPSC amplitudes was observed following local electrical stimulation, with the paired pulse ratio (PPR) unchanged after D3R agonist administration, suggestive of a postsynaptic response for D3R agonist. Additionally, cariprazine, a partial D3 agonist, mirrored the effect of the D3R full agonist.

In contrast, quetiapine, which acts as a D3R G-protein antagonist and beta-arrestin agonist, demonstrates an increase in the magnitude of evoked post-synaptic inhibitory currents in D3R-expressing neurons. Further investigation revealed that this effect is observed in D1+/D3+ double-positive cells but not in D1+ or D3+ single-positive cells. These results suggest that quetiapine's effects may involve combinatorial interactions between D1 and D3 receptors, and potentially non-G protein pathways, such as beta-arrestin signaling. Analysis of HCR imaging confirms significant overlap in D1R and D3R expression in nucleus accumbens medium spiny neurons.

Conclusions: By examining the specific function of D3R in the nucleus accumbens in mice we are able to better understand the action of currently prescribed SGAs and advance development of more efficacious treatments with less significant side effect burdens. Combining pharmacology and electrophysiological tools to study the local circuitry of the nucleus accumbens, we can begin to tease apart the physiological function of D3Rs in the ventral striatum and identify potential new targets for future therapeutics. Our results reveal the effects on inhibitory output from nucleus accumbens medium spiny neurons downstream of D3R action with different pharmacologic agents. We also demonstrate the further need in understanding the complexity in region-specific dopamine receptor pharmacology.

Keywords: Molecular Pharmacology, Dopamine (D2, D3) receptors, Nucleus Accumbens, G protein coupled receptors, β -arrestin

Disclosure: Nothing to disclose.

P139. ATYPICAL NUCLEUS ACCUMBENS NEURONS LOCALLY REGULATE DOPAMINE AND SHAPE OPIOID WITHDRAWAL AND RELAPSE BEHAVIORS

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Background: The opioid epidemic creates an urgent demand for effective treatments for opioid use disorder. The negative emotional state of withdrawal, along with environmental cues linked to opioid use, strongly contributes to relapse and elevates the risk of overdose. Although dopamine (DA) signaling in the nucleus accumbens (NAc) is known to increase during drug use and decrease during withdrawal, the precise neural circuits underlying these shifts remain unresolved and are the subject of ongoing debate. Finally, recent transcriptomic studies have uncovered greater cellular diversity within motivation-related circuits, suggesting that distinct cell types may play specific and critical roles across different stages of addiction and opioid use disorder.

Methods: We used female and male wild-type and transgenic mice to study cell-type-specific mechanisms in opioid use and withdrawal. Single-cell RNA sequencing of mouse NAc and human striatum identified distinct cell populations. Morphine induced dependence and withdrawal ($n = 10–20$), while neural activity was monitored with dopamine and calcium sensors ($n = 6–8$) and manipulated using opto- and chemogenetics ($n = 8–14$). Cre-mediated deletion of opioid receptors targeted specific neuronal types ($n = 10–16$). Withdrawal aversion was tested via naloxone-induced CPA ($n = 8–16$), and fentanyl self-administration, extinction, and reinstatement were paired with chemogenetic tools to probe behavioral regulation ($n = 8–10$). Samples sizes of 8–14 mice were used for behavior, optogenetic or chemogenetic manipulations, and 6–8 mice were used for in vivo monitoring with calcium or dopamine sensors. All studies were analyzed with unpaired, two-tailed-tests, two-way ANOVAs, repeated measures when appropriate, and mixed effect linear models when appropriate. A criterion of $p < 0.05$ was used for statistical significance.

Results: Morphine withdrawal produced robust behavioral aversion and a marked decrease in dopamine (DA) release within the nucleus accumbens (NAc). In vivo recordings confirmed strong inhibition of DA axon activity during withdrawal, independent of midbrain GABAergic inputs. Single-cell RNA sequencing identified a distinct NAc cell population co-expressing the highest transcript level of mu opioid receptor (Oprm1) and dopamine receptor D1 (Drd1), but largely lacking the canonical marker dynorphin (Pdyn). Instead, this population was defined by high expression of the transcription factor Tshz1 and was also observed in human striatal tissue. Tshz1 neurons were suppressed by morphine but exhibited rebound excitation during withdrawal. In naive mice, activating Tshz1 neurons strongly inhibited DA release in the NAc via local GABAergic signaling onto DA axons. Targeted chemogenetic inhibition or Oprm1 deletion in these cells prevented both withdrawal-induced aversion and DA suppression. Further, Tshz1 expressed a cellular specific, druggable GPCR that when agonized

led to reduced withdrawal aversion. Finally chemogenetic excitation of NAc Tshz1 neurons during fentanyl IV self administration led to accelerated extinction and reduced reinstatement with ongoing experiments delineating the effects of chemogenetic inhibition.

Conclusions: These findings reveal Tshz1 neurons as a conserved and functionally distinct D1 medium spiny neuron population that modulates DA signaling during opioid withdrawal and suggest a novel, cell-type-specific therapeutic target for alleviating withdrawal symptoms and reducing relapse risk.

Keywords: Mu opioid receptor, Medium Spiny Neuron, opioid withdrawal, opioid dependence, pharmacotherapy, animal model, withdrawal

Disclosure: Headlamp Health, Consultant, Self.

P140. CHARACTERIZATION OF THE CELL TYPES AND GENE EXPRESSION PATTERNS IN THE RHESUS MONKEY BASAL AMYGDALA USING SINGLE-NUCLEUS RNA SEQUENCING

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Background: The amygdala is a complex structure conserved across species and is a core component of the distributed neural circuit underlying threat and anxiety processing. Alterations in amygdala function are hypothesized to underlie pathophysiological processes relevant to anxiety and other stress-related disorders. Anxiety disorders emerge during the pre-adolescent and adolescent periods, which coincide with significant developmental changes in amygdala structure and reactivity. In the amygdala, the basal amygdala (BA) is strategically located to serve as an information processing hub and has been implicated in stress-related psychopathology. The evolutionary relatedness of rhesus monkeys to humans makes this non-human primate species invaluable for modeling human psychopathology to uncover underlying molecular and neural mechanisms. We have used young rhesus monkeys to study mechanisms underlying anxious temperament (AT), a phenotype that confers considerable risk of developing anxiety. In the present study, single-nucleus RNA-seq (snRNA-seq) was performed on BA tissue obtained from 72 rhesus monkeys that were phenotyped for AT, ranging in age from 1.8 to 4.3 years, an age range that in humans is comparable to pre-adolescence - adolescence. The goal was to describe the cell-type-specific gene expression patterns of the rhesus BA region. We also sought to understand the extent to which AT, age, and HPA axis activity were related to cell type-specific gene expression patterns.

Methods: Immediately before euthanasia, plasma was collected for measurement of cortisol, and then animals were deeply anesthetized with pentobarbital and euthanized for brain collection. Brain tissue was snap frozen in 3.5 mm thick coronal slabs and subsequently, tissue punches (3 mm diameter) corresponding to the BA region were collected. In the Binder laboratory, nuclei were extracted from ~20 mg of frozen tissue. For single-nucleus RNA sequencing (snRNA-seq), libraries were prepared using the 10x Genomics Chromium kit (v3.1), targeting 10,000 nuclei per sample. Libraries were sequenced on an Illumina NovaSeq X Plus System. Sequence reads were demultiplexed, aligned to the *Macaca mulatta* genome (NCBI 103), and processed using cell ranger count (Cell Ranger v8.0.1; 10x Genomics), including the argument --include-introns. Quality control excluded four samples, resulting in a final dataset of 745,497 nuclei from 68 subjects, with 22,270 expressed genes. Data were normalized using scran, followed by feature selection, dimensionality reduction, clustering, and cell type annotation. AT was assessed using the no-eye-

contact (NEC) condition of the human intruder paradigm (HIP) that consisted of a human intruder presenting their profile to the monkey without making direct eye contact. The AT score was calculated as a composite of freezing behavior and coo vocalizations during the NEC, and plasma cortisol values were assessed immediately after the NEC. Plasma cortisol concentrations were measured using a radioimmunoassay (MP Biomedicals, Solon, OH).

Results: Glial cell types (including astrocytes, ependymal cells, oligodendrocyte precursors, oligodendrocytes, and microglia) were annotated based on well-established marker genes from the literature. Neuronal cell types were assigned using previous studies conducted in the rhesus macaque amygdala and across species. Among the neuronal cell types, 73.1% were excitatory and 26.9% were inhibitory, aligning well with previous findings of a ratio of 70.2 to 29.8% in the basal amygdala. We identified 10 excitatory neuron clusters, some of which matched previously described cell type annotations. Inhibitory neuron types could be separated based on developmental origin, such as CGE-derived (ADARB2+, PROX1+, NR2F2+), LGE-derived (MEIS2+, ISLR2+), and MGE-derived (NKX2-1, LHX6, SOX6). Differential gene expression (DGE) analysis indicated that individual differences in cortisol levels had little effect on overall gene expression patterns. However, focused analyses on canonical genes involved in glucocorticoid signaling, such as FKBP5 and ZBTB16, revealed associations between gene expression and cortisol levels in several glial cell types. DGE analysis showed a total of 182 (unique) genes with age-related expression changes, most of which were downregulated with increasing age. These included EFNA5, a gene involved in synaptic plasticity, SYTL5, which is relevant for synaptic vesicle trafficking and neurotransmitter release, as well as SOX4, a transcription factor involved in neural development. Finally, no significant correlations were found in relation to AT.

Conclusions: We used snRNA-seq to characterize the cell type composition and gene expression patterns of the BA region in a large sample of young rhesus monkeys. We also identified between-subject differences in gene expression patterns that are correlated with age and with HPA axis activity. No significant gene expression patterns were identified as being associated with AT. Future studies will incorporate bulk RNA-seq data obtained from the same BA samples, which will complement the snRNA-seq by providing a greater depth of sequencing with enhanced sensitivity to detect less prominent transcripts. Beyond the snRNA-seq data, the analysis of bulk sequencing data has the potential to identify AT-associated genes with implications for understanding anxiety-related pathophysiology and for identifying novel anti-anxiety treatment targets.

Keywords: snRNASeq, Nonhuman Primates, Anxiety, FKBP5, basolateral amygdala

Disclosure: Nothing to disclose.

P141. THE ALCOHOL METABOLITE ACETATE IMPACTS THE EPIGENOME AND GENE EXPRESSION IN A SEX-, CONTEXT-, AND BRAIN REGION-SPECIFIC MANNER

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Background: Acetate derived from alcohol is directly deposited on histones in the brain, thus regulating gene expression and key behavioral outcomes such as the encoding of alcohol-induced associative memories. The specific epigenetic modifications

influenced by acetate, and the associated gene expression programs, however, remain poorly understood.

Methods: We used liquid chromatography coupled with tandem mass spectrometry to identify the effect of acetate on histone post-translational modifications in key brain regions following i.p. exposure to alcohol or acetate. RNA sequencing was used to assess effects on gene expression. Both sexes were studied ($n = 4$ per sex per group).

Results: We showed that acetate is deposited onto histones in a time- and dose-dependent manner, and increased the acetylation of both canonical histones and histone variants. Intriguingly, this epigenetic impact was sex-, brain region- and context-specific. Moreover, acetate led to the upregulation of activity-dependent genes and genes previously linked to memory formation in the dorsal hippocampus of female mice.

Conclusions: Overall, our findings shed new light onto key epigenetic and transcriptional mechanisms influenced by the alcohol metabolite acetate, which play a critical role in the formation of alcohol-associated memories and the development of alcohol use disorders. Thus, our results will guide the development of novel therapeutic interventions.

Keywords: Drug metabolism, Epigenetics, gene expression

Disclosure: Nothing to disclose.

P142. SUPPRESSION OF INTERLEUKIN-33 SIGNALING IN A MOUSE MODEL OF ALCOHOL DEPENDENCE

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Background: Recent work in our lab indicates that the intrinsic excitability of nucleus accumbens (NAc) neurons is suppressed during early withdrawal in a mouse model of alcohol dependence. Moreover, NAc neuronal excitability is negatively correlated with the level of voluntary alcohol drinking. We hypothesized that this alcohol-related NAc neuroplasticity involves neuroimmune signaling and glia-neuron crosstalk. Interleukin-33 (IL-33) is a multi-functional cytokine implicated in peripheral consequences of heavy alcohol consumption, and its expression in the NAc is decreased in a mouse model of genetic risk for excessive drinking. Pilot studies established that, in the NAc, astrocytes produce IL-33 and neurons are functionally responsive to it. Here, we assessed how NAc IL-33 signaling and its effects on neuronal function are impacted by alcohol dependence.

Methods: Male and female mice were treated with 2-bottle choice (water versus 15% ethanol) self-administration in combination with cycles of intermittent air (control mice), or ethanol vapor exposure and withdrawal (to induce dependence). Tissue was collected for molecular assessments immediately after the final vapor session (total $N = 71$ (35 males, 36 females); $n = 11$ –12 mice per ethanol treatment group per sex). RNA-Seq of NAc tissue punches was performed on bulk input and on an astrocyte RNA enriched fraction, and data was analyzed aggregated by sex, comparing dependent versus non-dependent treatment groups. Brain slices were prepared for slice electrophysiology 24 h after the final vapor session (total N (mice) = 62 (28 males, 34 females). Whole-cell patch clamp recordings of passive and active membrane properties were collected from NAc neurons during incubation with 0, 10, or 100 ng/mL IL-33 ($n = 7$ –19 neurons from 8 to 12 mice per IL-33 concentration per ethanol treatment group per sex). Excitability measures were analyzed separately for males and females by 2-way ANOVA with IL-33 Concentration and Ethanol Treatment Group as factors.

Results: (a) Astrocyte IL33 was decreased ($\log_2\text{foldchange} = -0.28$, $p\text{-adj} = 0.039$) in dependent mice relative to non-dependent mice (which had a non-significant reduction relative to ethanol naïve). Ptk2 (encoding a positive regulator of IL33 transcription) and downstream signaling molecules Myd88 and Stat3 were also differentially expressed in dependent versus non-dependent mice (Ptk2: $\log_2\text{foldchange} = -0.21$, $p\text{-adj} = 0.045$; MyD88: $\log_2\text{foldchange} = 0.45$, $p\text{-adj} = 0.027$; Stat3: $\log_2\text{foldchange} = 0.43$, $p\text{-adj} = 0.0002$). (b) For males, maximum action potential firing frequency showed a main effect of IL-33 Concentration ($F(2, 85) = 3.1$, $p = 0.049$; Tukey's post-hoc comparisons: in non-dependent mice, 10 ng/mL > 0 ng/mL IL-33, $p = 0.003$). For females, action potential threshold differed as an interaction of IL-33 Concentration with Ethanol Treatment Group ($F(4, 109) = 3.5$, $p = 0.01$; Tukey's post-hoc comparisons: in non-dependent mice, 10 ng/mL < 0 ng/mL IL-33, $p = 0.004$, in dependent mice, 100 ng/mL > 0 ng/mL, $p = 0.048$).

Conclusions: Ethanol dependence reduced transcript levels of IL33 and a positive regulator of its transcription Ptk2, specifically in astrocytes, while downstream signaling molecules MyD88 and Stat3 were upregulated. The specific physiological measures altered by IL-33 incubation differed between females and males, but for both sexes, 10 ng/mL IL-33 promoted excitability of neurons from non-dependent mice, while neurons from dependent mice only responded to 100 ng/mL. These results indicate that NAc IL-33 production and/or signaling by astrocytes is dysregulated in alcohol dependence, while sensitivity to IL-33-mediated stimulation of neuronal activity is lost. Given that we find a negative relationship between voluntary alcohol intake and NAc neuronal excitability, these findings suggest that dysregulation of IL-33 signaling may promote escalation of drinking by dependent mice.

Keywords: astrocyte-neuron interaction, slice electrophysiology, cytokine, neuroimmune mechanisms, Alcohol dependence

Disclosure: Nothing to disclose.

P143. TARGETING THE GUT MYCOBIOME-IMMUNE AXIS WITH PACHYMAN REVERSES PSYCHOSOCIAL AND COGNITIVE DEFICITS IN ECOHIV-INFECTED FEMALE MICE

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Background: HIV-associated psychosocial and cognitive deficits persist despite antiretroviral therapy, underscoring the need for new mechanisms and interventions. Emerging data implicate gut fungi (the mycobiome) in neuroimmune signaling, but their role in HIV-related behavioral impairment is unclear. We tested whether a mycobiome-immune axis links to EcoHIV-induced deficits in female mice and whether targeting this axis with the beta-glucan pachyman is therapeutic.

Methods: Female C57BL/6 mice were infected with EcoHIV and treated with pachyman or vehicle. Behavior was assessed by open field, elevated plus maze, three-chamber social interaction, and novel object recognition. Colonic lamina propria immune cells were analyzed by flow cytometry. Gut mycobiome profiles were generated via ITS2 rRNA gene sequencing, and metabolic pathway alterations were assessed by untargeted metabolomics.

Results: EcoHIV infection was associated with a marked increase in the fungal species *Malassezia globosa* that correlated with impaired social interaction. Pachyman significantly reversed

social avoidance and cognitive deficits ($P < 0.01$) without affecting locomotor or anxiety-like behaviors. EcoHIV increased colonic Dectin-1+ macrophages ($P < 0.01$), and pachyman reduced this change ($P < 0.05$). Dectin-1+ macrophage levels positively correlated with social behavior disruption ($r = 0.5209$, $P = 0.008$) and with fungal abundance ($r = 0.5285$, $P = 0.0074$). EcoHIV altered key immune-regulatory metabolic pathways, which were restored by pachyman.

Conclusions: Disruption of the gut mycobiome-immune axis contributes to EcoHIV-induced psychosocial and cognitive impairments in female mice. Targeting this axis with pachyman reversed behavioral deficits and reduced colonic Dectin-1+ macrophage activation, supporting fungal-immune modulation as a therapeutic strategy for HIV-associated neuropsychiatric and cognitive disorders.

Keywords: Mycobiome, Dectin-1, EcoHIV, Psychosocial behavior, Pachyman

Disclosure: Nothing to disclose.

P144. PROTEOMIC PROFILING OF MONOAMINERGIC AXONS

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Background: A common anatomical feature of monoamine projection neurons is their extensive, unmyelinated axons that project to the forebrain which may underlie their selective vulnerability across brain regions and diseases. Traditional methods to overcome the limitations of bulk tissue proteomics include single-cell sorting and laser capture microdissection, but these methods cannot capture fine neuronal processes such as dendrites and axons. The ideal analysis of presynaptic proteomic architecture requires direct measurement of protein levels within the axon itself. We have recently developed a proximity-labeling proteomics protocol employing cytoplasmic APEX2 that enables axon-specific proteomic profiling of projection neurons, which allows for high resolution mapping of the axonal and presynaptic proteome in native brain tissue. Here, we apply this method for comparative proteomic profiling of dopaminergic and serotonergic axons in the mouse forebrain.

Methods: To express cytoplasmic APEX2 in dopaminergic or serotonergic neurons, we employed a Cre-dependent AAV5 viral vector that expresses V5-tagged APEX2 fused to a nuclear export sequence (NES). Injection of AAV5-CAG-DIO-APEX2NES into the ventral midbrain of DAT-IRES-Cre mice or into the dorsal raphe of SERT-Cre mice produced robust APEX2 expression specific to the targeted neurons. Both male and female mice were used in these studies. Axonal APEX2 labeling was conducted in acute slices, with key steps including: (1) transcardial perfusion with low-sodium cutting solution, (2) incubation of slices with BP in artificial cerebrospinal fluid (aCSF), (3) rapid labeling with H₂O₂ in aCSF (3 min), and (4) rapid quenching with an antioxidant aCSF. Alternatively, whole mouse forebrains were directly homogenized under gentle conditions to isolate synaptosomes, which were directly labeled in vitro under conditions analogous to cell suspensions. The synaptosome approach is more rapid and less labor intensive, but still permits direct interrogation of the proteome derived from presynaptic terminals in the mouse brain.

Biotinylated samples were lysed and targeted protein capture was conducted using streptavidin beads. No-APEX2 samples were employed as controls for non-specific binding. On-bead digestion was conducted and quantitative mass spectrometry-based proteomics was carried out using data-dependent acquisition (DDA). Total peptide- and protein-normalized abundances were analyzed

using differential expression analysis across sample groups using Welch's t-test with Benjamini-Hochberg procedure to control the false discovery rate (FDR). Log₂ transformation was employed for protein data visualization and clustering.

Results: We demonstrate that axonally expressed APEX2 proximity labeling proteomics can identify and quantify thousands of proteins per sample, and facilitate cross-neuron comparisons. We identified key elements of the presynaptic active zone release machinery in monoaminergic neurons, as well as numerous autoreceptors and heteroreceptors. We also obtained extensive coverage of proteins involved in synaptic vesicle trafficking, vesicle fusion, and synaptic vesicle priming. Overlapping and unique proteins were identified across dopaminergic and serotonergic axons, with clear differences in neurotransmitter synthesis and reuptake machinery. In dopaminergic axons, we identified an axonal enrichment of proteins encoded by Parkinson's disease-linked genes, with functional implications for PD pathophysiology.

Conclusions: Our study demonstrates APEX2 labeling and mass spectrometry-based proteomics of axonal and presynaptic compartments of genetically targeted monoaminergic neurons in the mouse brain. Thus, APEX2 labeling in acute brain slices provides a general approach for cell type-specific and subcellular-targeted proteomics in the mouse brain. Future studies will address the physiological role of identified autoreceptors and heteroreceptors in monoaminergic axons using optical techniques to measure neurotransmitter release.

Keywords: Dopamine, Serotonin, axons, Neurotransmitter release, Proteomics

Disclosure: Nothing to disclose.

P145. R-LOOPS, DNA:RNA HYBRID TRIPLE-STRANDED GENOME STRUCTURE, CONTROL GENE EXPRESSION AND REGULATE BEHAVIORAL PLASTICITY

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Background: Emotional experiences often drive adaptive behavioral responses, enabling organisms to respond flexibly to their environments. These experiences activate neuronal circuits and initiate the expression of activity-dependent genes, which are critical for cellular, synaptic, and behavioral plasticity. Dysregulation of these transcriptional programs contributes to maladaptive behaviors associated with neuropsychiatric disorders, including mood and substance use disorders (SUD). The expression of these genes is tightly regulated by complex intracellular signaling cascades and epigenetic mechanisms acting on genomic enhancers, which can be located far from the gene's proximal promoter. However, the precise mechanisms governing this regulation remains elusive. Long-noncoding RNAs (lncRNAs) have emerged as key regulators of gene expression through diverse mechanisms. One notable mechanism involves the formation of R-loops, three-stranded structures composed of a DNA:RNA hybrid and a displaced single-stranded DNA. R-loops are known to influence various transcriptional, translational, and DNA repair. However, the physiology and function of R-loops in the brain is only just beginning.

Methods: Adult male and female C57BL/6J mice were subjected to cocaine conditioned place preference (CPP) paradigm or a single social defeat stress (SDS). To manipulate lncRNA and R-loop, mice received injections of short-hairpin RNA (shRNA) targeting lncRNAs, lncRNA overexpression vectors, or a dCas9-

fused with RNaseH1 (CRISPR-H1) which selectively degrade DNA:RNA R-loop at targeted genomic locus using single-guide RNA in the medial prefrontal cortex (mPFC) or nucleus accumbens (NAc). Treated mPFC and NAc tissues were collected immediately after behavioral stimuli for subsequent molecular analysis. Gene expression was examined using RNA-seq and qPCR ($n = 7-12$). R-loop formation was assessed by DNA:RNA hybrid immunoprecipitation (DRIP) followed by qPCR ($n = 7-20$) or sequencing. Chromatin looping was examined using chromosome conformation capture (3C) ($n = 8-20$). Mice received shRNA or dCas9-RNaseH1 were exposed to chronic SDS, followed by behavioral testing, including the sucrose preference assay for depression-like behaviors ($n = 12-27$) and cocaine CPP test for SUD-related behaviors ($n = 10-12$). Statistical analyses included One-Way and Two-Way ANOVA with Tukey's, Sidak's, or Bonferroni's post-hoc comparison. Significance was considered $p < 0.05$.

Results: RNA-seq analysis identified lncRNAs transcribed from enhancers of activity-dependent genes, including *Npas4* and *Fos* in the NAc and mPFC. These lncRNAs formed R-loops at enhancers, with R-loop formation dynamically regulated in a genomic locus-specific manner in response to emotional stimuli, such as novel environment exposure or SDS ($p < 0.05$, $n = 7-20$). Disruption of R-loops using CRISPR-H1 significantly reduced *Npas4* expression ($p < 0.01$, $n = 5-10$), demonstrating their essential role for activity-dependent gene expression. 3C analysis showed that emotional stimuli enhanced enhancer-promoter interaction at the *Npas4* gene, and degradation of R-loops abolished this chromatin looping ($p < 0.05$, $n = 8-10$). Behaviorally, mice with disrupted R-loops at the *Npas4* enhancer failed to develop cocaine CPP showed that R-loops are essential for the development of cocaine reward associated contextual memory ($p < 0.05$, $n = 10-12$). Genome-wide R-loop profiling revealed widespread R-loop formation, with enrichment at the genes involved in long-term potentiation, glutamate and dopamine signaling, and calcium signaling.

Conclusions: Our study identified lncRNAs expressed at enhancer regions of activity-dependent genes, such as *Npas4* and *Fos*, in the NAc and mPFC. These lncRNAs form R-loops in a neuronal activity-dependent and genomic locus-specific manner in response to emotional experiences. R-loops at the *Npas4* enhancer regulate the interaction between distal enhancer and proximal promoter through 3D chromatin looping, and modulating activity-dependent gene expression, and contributing to the formation of cocaine reward-associated contextual memory. Our genome-wide analysis reveals that R-loops are enriched at genes involved in synaptic plasticity. These findings suggest that the R-loops represent a novel epigenetic regulatory mechanism in the transmission of emotional experience into the gene expression underlying behavioral adaptations. At this conference, we will discuss the molecular mechanisms and functional significance of R-loops in the synaptic and behavioral plasticity.

Keywords: DNA:RNA hybrid R-loop structure, long noncoding RNA, long-range chromatin interaction, *Npas4*, Adaptive Behavior

Disclosure: Nothing to disclose.

P146. METHAMPHETAMINE-INDUCED HYPOFRONTALITY: NPAS4 AND THE NEURAL BASIS OF COGNITIVE DEFICITS

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Background: Hypofrontality, a pathological state marked by reduced function of the medial prefrontal cortex (mPFC) circuits, is

a shared neurobiological feature across various neuropsychiatric disorders, including substance use disorder, schizophrenia, depression, and ADHD. This condition is characterized, in part, by a disruption of the excitatory and inhibitory (E-I) synaptic transmission balance within the PFC. Individuals with hypofrontality often exhibit cognitive and executive dysfunctions, such as working memory deficits, attention problems, and increased impulsivity. Among substances known to contribute to hypofrontality, methamphetamine (METH) is of particular concern due to its potent and long-lasting neurobehavioral effects. However, the molecular and cellular mechanisms driving the METH-induced E-I imbalance and cognitive dysfunction remain poorly understood. This project focuses on neuronal PAS domain protein 4 (NPAS4), an activity-dependent immediately early gene known to regulate E-I balance in a cell-type-specific manner. We aim to investigate the functional role of NPAS4 in METH-induced impairment in E-I transmission and the role of NPAS4 expressing neuronal ensembles in cognitive dysfunctions associated with METH exposure.

Methods: Cohorts of young adult C57BL/6J mice underwent a repeated methamphetamine (METH) protocol. Mice received METH on days 1 and 7 (1.0 mg/Kg, i.p.) and daily from days 2–6 (3.0 mg/kg, i.p.). Following 7 days of home-cage abstinence, animals were tested for working memory, anxiety-like behavior, and exploratory activity. In separate cohorts that underwent the same METH sensitization and abstinence protocol, ex vivo whole-cell patch-clamp recordings were performed in the prefrontal cortex (PFC) to assess GABAergic transmission. Additionally, NPAS4 expression in the PFC was examined via immunohistochemistry (IHC) following acute METH injections and after repeated METH exposure (same protocol as above), followed by abstinence periods of 1 h, 24 h, or 7 days.

Furthermore, in a cohort of rats treated with saline ip (14 days) or METH (1.0 mg/Kg days 1 and 14; 5.0 mg/Kg days 2–13), followed by 7 days of home cage abstinence, adeno associated virus (AAV)-mediated expression of a short-hairpin RNA targeting *Npas4* (sh*Npas4*) was utilized to reduce *Npas4* expression in the mPFC of rats and working memory was assessed.

Results: Behavioral assessments in mice show that METH exposure is associated with a significant increase in anxiety-like behavior, as indicated by reduced time spent in the open arms of the Elevated Plus Maze (SAL: 8.3 ± 1.8 s, $n = 25$; METH: 3.7 ± 0.7 s, $n = 22$; t-Test $p < 0.003$), and a significant decrease in center time during the Open Field test (SAL: 55.01 ± 1.3 s, $n = 37$; METH: 33.8 ± 3.2 s, $n = 30$; t-Test $p < 0.0001$).

Rats treated with shRNA NPAS4, showed a significant increase in the score for the delay no match to sample test (DNMS) 2-way ANOVA $p < 0.01$.

Electrophysiological recordings from the prefrontal cortex (PFC) of mice, show that repeated METH administration significantly increase sIPSC amplitude (SAL: 17.6 ± 8.7 pA, $n = 10$; METH: 27.8 ± 6.3 pA, $n = 23$; t-Test $p < 0.006$) and significantly reduce the intrinsic excitability of glutamatergic pyramidal neurons (t-Test $p < 0.0001$).

Immunohistochemical analysis reveal that a single acute dose of METH (5.0 mg/kg) significantly increases the number of cells expressing NPAS4 (t-Test $p < 0.0003$) in the PFC one-hour post-injection. Furthermore, the increase in neurons expressing NPAS4 was sustained following 7 days of injections ($p < 0.001$) and begin to decline after 24 h of abstinence. Furthermore, METH induced a dose-dependent increase in NPAS4+ cell counts within the PFC. Notably, approximately 95% of NPAS4+ cells colocalize with CaMKII-expressing neurons, with significantly greater induction observed in the prelimbic (PrL) region compared to the infralimbic (IL) PFC.

Conclusions: Our findings demonstrate that repeated METH administration induces significant cognitive deficits in mice and rats, accompanied by enhanced inhibitory synaptic activity in the

prefrontal cortex (PFC) and robust upregulation of the early immediate gene NPAS4. These results highlight a potential molecular and circuit-level mechanism underlying METH-associated hypofrontality and cognitive dysfunction.

Keywords: Methamphetamine, Hypofrontality, Npas4, Medial Prefrontal Cortex (mPFC), mice

Disclosure: Nothing to disclose.

P147. INTEROCEPTIVE NETWORK RESPONSES TO INFLAMMATORY CHALLENGE IN DEPRESSION

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Background: Peripheral inflammation generates interoceptive signals that communicate changes in visceral and somatic states to the brain, driving adaptive shifts in motivation and energy conservation strategies collectively known as “sickness behavior”. These actions are likely mediated by a brain network which maps bodily signals onto networks representing the internal state (including cortical hubs such as the insular cortex) and then integrating these signals with affect, self-awareness, and adaptive behavioral regulation (including the cingulate cortex). Although circulating inflammatory mediators are elevated in a subgroup of individuals with major depressive disorder (MDD), it remains unclear whether interoceptive circuits process these immune signals differently in depressed compared to healthy individuals.

Methods: Twenty-one adults with MDD and eight healthy comparisons (HC) received low-dose lipopolysaccharide (LPS; 0.8 ng/kg). Functional MRI was performed before (baseline scan) and ~2 h post-LPS infusion, coinciding with approximate peak of the inflammatory cytokine response. During scanning, participants completed the visceral interoceptive awareness task in which an interoceptive condition (attention to heart/stomach sensations) was contrasted with an exteroceptive control condition (attention to font-color changes). Serum cytokines were measured using MesoScale Discovery V-Plex assays. Human immune-challenge studies (endotoxin; typhoid vaccine) consistently implicate insula and cingulate activation and connectivity in cytokine-linked changes in mood, fatigue, and social-affective processing. Therefore, our analyses tested diagnosis by time interaction effects within insula and cingulate cortex subregions defined by the Brainnetome atlas, controlling for age, sex, BMI and in-scanner head motion.

Results: LPS induced robust inflammatory serum cytokine responses in both groups, peaking at 1.5 h post infusion. In HC, LPS-induced inflammation was accompanied by bilateral decreases in activation of both posterior insular (i.e., hypergranular and dorsal granular subregions) and posterior cingulate cortex (i.e. ventral and dorsal area 23) during interoceptive attention. In contrast, participants with MDD showed no reductions and in some cases exhibited increased signal changes in these same regions. The 2 (Diagnosis: MDD, HC) × 2 (Time: pre- vs post-LPS) mixed-effects model revealed significant Diagnosis × Time interactions in both regions (posterior insula, $d = 0.74-0.78$; posterior cingulate, $d = 0.97-1.06$; all $p_{FDR} < 0.05$), indicating larger post-LPS signal decreases in HC relative to MDD.

Conclusions: These preliminary findings suggest that MDD may be characterized by aberrant neural representation of immune signaling in posterior insula and posterior cingulate in the context of an inflammatory challenge. Future work should examine whether this potentially altered central registration of peripheral inflammation hinder the resolution of immune responses and

predict the persistence of depressive symptoms in the context of inflammation. Confirming these observations in larger samples would provide valuable information about how altered immune-brain communication via interoceptive networks might represent a mechanistic pathway linking inflammation to depression.

Keywords: Depression, Task-based fMRI, neuroimmune mechanisms, endotoxin, interoception

Disclosure: Nothing to disclose.

P148. A PIVOTAL PHASE 3 SAFETY TRIAL OF MULTIPLE DOSING WITH BXCL501 (SUBLINGUAL DEXMEDETOMIDINE) FOR TREATMENT OF AGITATION ASSOCIATED WITH BIPOLAR DISORDER OR SCHIZOPHRENIA IN THE HOME SETTING

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Background: BXCL501 is a sublingual film formulation of dexmedetomidine, a selective alpha-2 adrenergic receptor agonist, which has been approved for treatment of episodes of agitation in patients with schizophrenia or bipolar disorder under the supervision of a healthcare provider. The only other currently approved therapies for management of acute agitation in these patients are injectable or inhaled formulations of antipsychotics. None of these medications are appropriate or approved for the home setting. However, most episodes of agitation occur at home. Treating episodes of agitation before they escalate could provide timely relief to patients and caregivers and reduce the need for emergency interventions or hospitalizations. In order to address this patient need, a Phase 3 safety trial of the lower of the two approved doses of BXCL501 (120 mcg) was initiated following consultations with the FDA. The primary objective of the trial was to investigate whether patients could safely self-administer BXCL501 in the absence of a healthcare provider. In addition, previous pivotal trials with sublingual dexmedetomidine film were conducted with single doses and there are limited data on whether repeated frequent dosing would continue to provide treatment benefit.

Methods: A randomized, double-blind, placebo-controlled, Phase 3, 12-week trial (SERENITY At-home, NCT05658510) was designed to evaluate the safety and tolerability of a 120 mcg dose of BXCL501 in adult patients with episodes of agitation associated with a primary diagnosis of bipolar disorder or schizophrenia in the community (at home) setting. Inclusion criteria included history of agitation episodes in the 3 months preceding enrollment and receiving stable psychotropic treatment for at least 30 days prior to enrollment. Exclusion criteria included unstable medical illness, history of agitation due to substance abuse, and history of syncope, hypovolemia, or clinically relevant ECG abnormalities. Patients living alone, as well as those who had a caregiver or another person spending significant time with them (informant) were enrolled. Patients were recommended to take the study product when they felt they needed it for management of agitation and were also allowed to employ alternative interventions at any point during the episode (meditation, alcohol, cannabis, etc.) in order to reflect real-world coping strategies.

The primary outcome measure of the trial was incidence of serious adverse events (SAEs) and treatment-emergent adverse events (TEAEs), defined as adverse events occurring within 24 h of dosing. In addition, exploratory outcome measures included a modified clinical global impression – severity (mCGI-S) in order to evaluate the benefit of BXCL501 over the course of the trial. mCGI-

S was scored by patients themselves and by caregivers/informants, if present, on a scale of 0 (no agitation) to 3 (severe agitation)

The sample size of 200 patients with at least one treated agitation episode, 100 per treatment arm, was believed sufficient to provide information for assessing adverse drug reactions that might occur in the at-home use setting.

Results: A total of 246 patients were enrolled in the trial, of which 31 did not experience any agitation episodes and 7 did not use investigational treatment for their episodes. Overall, 2437 agitation episodes were treated in 208 patients.

In this study, sublingual dexmedetomidine was generally well tolerated – there were no discontinuations due to drug-related adverse events and there were no treatment-related SAEs. Most TEAEs were mild. The most common TEAE was somnolence, occurring in 22.5% of patients with the first dose, similar to the occurrence of the same TEAE in the previous single-dose pivotal studies in the institutional setting (22%). The overall tolerability was also similar between the first dose in this trial and the single dose in the previous trials. With repeat dosing, the incidence of TEAEs decreased, and across all dosed episodes only the rates of somnolence (13.9%) and dry mouth (4.8%) were higher than 2%.

In order to evaluate the continued benefit of BXCL501 treatment with repeat administration, patients (and informants, where available) recorded the severity of symptoms using mCGI-S at baseline and 2 h after administration. Using this exploratory measure, a greater proportion of patients experienced full resolution of symptoms with BXCL501 compared with placebo across the 2437 episodes regardless of severity at baseline. The reduction of symptoms with BXCL501 remained similar between earlier and later dosed episodes, indicating no attenuation of benefit with repeat dosing.

Conclusions: In this pivotal safety study, as-needed doses of sublingual dexmedetomidine 120 mcg were safe, well tolerated, and reduced the severity of agitation in subjects with schizophrenia or bipolar disorder over a 12-week treatment period when administered on an as-needed basis. Over repeat dosing, no increase in TEAEs was observed, and the reduction in symptoms remained consistent throughout the duration of the trial.

Keywords: Acute agitation, Dexmedetomidine, Bipolar Disorder, Schizophrenia (SCZ)

Disclosure: BioXcel Therapeutics, Inc., Employee, Self.

P149. HARNESSING ARTIFICIAL INTELLIGENCE TO IMPROVE DATA QUALITY CONTROL IN MULTI-SITE CLINICAL TRIALS FOR ENX-102 IN GENERALIZED ANXIETY DISORDER

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Background: ENX-102 is a subtype-selective GABA-A $\alpha_{2,3,5}$ positive allosteric modulator in development for Generalized Anxiety Disorder (GAD). In multi-site psychiatric trials, high-quality and reliable data collection is essential, yet clinician-administered outcome measures such as the Hamilton Anxiety Rating Scale (HAM-A) are susceptible to variability due to differences in rater skill, subjective interpretation, and rater drift over time. Centralized rater surveillance programs, in which independent reviewers re-rate audio-recorded interviews and provide feedback, are often used to mitigate these challenges. While effective, these programs are resource-intensive and can be difficult to scale in large, decentralized trials. Scalable solutions that preserve data quality without significantly increasing operational burden are needed.

Artificial Intelligence (AI), particularly large language models (LLMs), may provide a scalable supplement by generating standardized secondary scores for clinical interviews that could be used to help identify scoring discrepancies and flag potential quality concerns. This proof-of concept study evaluated whether LLMs could be used to produce parallel HAM-A scores from trial recordings and serve as a scalable supplement to traditional rater surveillance.

Methods: The ENCalM trial was a 9-week, multi-center, randomized, double-blind, placebo-controlled Phase 2 study designed to investigate the efficacy and safety of ENX-102 for the treatment of GAD (NCT05749055) at 31 sites in the United States. The primary endpoint was the HAM-A, administered by site raters through the Structured Interview Guide for the Hamilton Anxiety Rating Scale (SIGH-A). All SIGH-A interviews audio-recorded through a compliant and validated electronic Clinical Outcome Assessment (eCOA) platform. A rater surveillance program was implemented by the eCOA vendor (EMA-Wellness), in which independent expert clinicians reviewed and scored all screening and baseline interviews, as well as a sample of in-study interviews. For the AI evaluation, a set of 322 interviews with paired site rater and independent reviewer scores was selected. The process followed four stages: 1. Transcription: recordings were transcribed in the eCOA platform using a Microsoft Azure LLM optimized for speech-to-text conversion. 2. Initial scoring: a private GPT-4o LLM hosted on the eCOA platform's Azure server was used for the initial scoring model. The LLM was given inputs consisting of study-specific rater training materials and the SIGH-A scoring framework, then generated outputs that included item-level scores with accompanying explanatory rationales for an initial set of 24 interview transcripts. These AI-derived scores were compared with both site rater and independent reviewer scores. 3. Refinement: An expert clinician reviewed the outputs and provided targeted feedback to improve prediction performance, which were provided as instructional prompts to the LLM. The refined scoring model was then re-applied to the same subset of 24 interview transcripts. 4. Validation: the refined model was applied to the remaining set of 298 interviews to evaluate its performance more broadly.

Results: Across the full set of 322 interviews, site rates and independent reviewers demonstrated strong agreement, with a Pearson intraclass correlation coefficient (ICC) of 0.93 ($\chi^2 = 9529$, $p < 0.0001$). The initial AI scoring model showed moderate agreement with both site raters (ICC = 0.76, $\chi^2 = 3347$, $p < 0.0001$) and independent reviewers (ICC = 0.77, $\chi^2 = 3525$, $p < 0.0001$). After expert clinician feedback and refinement, model performance improved, with AI scoring showing higher ICCs with site raters (ICC = 0.79, $\chi^2 = 3693$, $p < 0.0001$) and independent reviewers (ICC = 0.80, $\chi^2 = 3936$, $p < .0001$). The aggregate mean difference between raters across items improved to 0.02 from 0.18 for site raters and 0.14 for reviewers. Finally, when the refined model was applied to the broader interview set, it maintained good agreement with both site raters (ICC = 0.76, $\chi^2 = 3959$, $p < 0.0001$) and independent reviewers (ICC = 0.77, $\chi^2 = 4175$, $p < 0.0001$). The aggregate mean scoring discrepancy between AI rating and both other sets of raters was < 0.01 , supporting its ability to generalize beyond the training sample.

Conclusions: This proof-of-concept study supports the feasibility of using LLMs to augment rater surveillance in multi-site psychiatric trials that rely on clinician-rated outcomes such as the HAM-A. The AI model showed promising agreement with human raters and benefitted from targeted expert input to improve scoring performance. Future work will involve further refining the model through additional clinician feedback and validation using larger datasets. LLMs have the potential to enhance the efficiency and scalability of centralized quality control by detecting rater drift, flagging anomalous scores, and providing real-time feedback. The refined model derived from the ENX-102 Phase 2 data

may serve as a valuable supplemental tool for ensuring data quality in the Phase 3 program in GAD.

Keywords: Generalized Anxiety Disorder, Artificial Intelligence, Data Quality Monitoring

Disclosure: Engrail Therapeutics, Employee, Self.

P150. FEASIBILITY AND PRELIMINARY EFFICACY OF TIRZEPATIDE FOR INDIVIDUALS WITH MODERATE OR SEVERE METHAMPHETAMINE USE DISORDER

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Background: Glucagon-like peptide-1 (GLP-1) receptor agonists have gained recent attention for drug repurposing to treat substance use disorder. Yet, tirzepatide, a dual agonist of GLP-1/glucose-dependent insulinotropic polypeptide, has not been studied systematically in individuals with substance use disorders, especially for methamphetamine use disorder.

Methods: Individuals with moderate or severe methamphetamine use disorder who met the FDA-approved indication of tirzepatide for weight management were enrolled in an ongoing open-label trial of 36-week duration. This interim analysis included data up to 16 weeks and was conducted to provide evidence for feasibility and efficacy to inform larger-scale multi-site double-blind randomized controlled trial. Descriptive statistics were used to describe the baseline clinical and demographic variables. Repeated measured mixed model analyses with week as the within subject factor were used with unstructured covariance. Separate mixed model analyses with week as the independent variable of interest and age and sex as covariates were used for the following dependent variables: (1) craving over the past week that was assessed using a Visual Analog Scale (VAS; 0 = did not crave methamphetamine at all, 100 = had the most intense craving possible); (2) current craving that was assessed using 10-item Stimulant Craving Questionnaire (STCQ); (3) overall depression using the Quick Inventory of Depressive Symptoms (QIDS-SR); and (4) impulsivity with Concise Health Risk Tracking (CHRT) scale. Least square (LS) means estimates were obtained for change at each week compared to baseline.

Results: Of the 42 individuals screened, 35 were enrolled [20 females/15 males; mean (standard deviation, SD) age = 46.1 (8.3) years]. Two individuals dropped out after the first dose and one individual dropped out after the seventh dose due to gastrointestinal side effects. Overall adherence to weekly injection visits was 92.3%. Mean (SD) of body mass index, weight (in pounds), VAS, STCQ, QIDS-SR, and CHRT impulsivity at baseline were 35.9 (7.5), 224.8 (50.7), 57.0 (24.8), 41.6 (12.4), 8.2 (4.4), and 4.5 (2.0), respectively. After 16 weeks of treatment with tirzepatide, there were significant reductions in past week craving (VAS) for methamphetamine [estimate(β) = -18.9, standard error(SE) = 4.2, $p < 0.0001$], current craving (STCQ) for methamphetamine (β = -11.9, SE = 2.0, $p < 0.0001$), depression (QIDS-SR) (β = -3.2, SE = 0.6, $p < 0.0001$), and impulsivity (CHRT) (β = -1.6, SE = 0.3, $p < 0.0001$). Higher levels of depression and impulsivity were associated with higher cravings for methamphetamine.

Conclusions: This is the first study of a GLP-1 receptor agonist in individuals with methamphetamine use disorder. Furthermore, this is the first study to use tirzepatide and evaluate its effects on substance use-related outcomes in any substance use disorder. This study supports the feasibility of using tirzepatide for

methamphetamine use disorder, and provides preliminary evidence for its efficacy in methamphetamine use disorder by reducing craving for methamphetamine, depression, and impulsivity. Larger-scale double-blind randomized controlled trials are needed to evaluate the therapeutic utility of tirzepatide for methamphetamine use disorder.

Keywords: Tirzepatide, Methamphetamine, craving, impulsivity, Depression

Disclosure: Janssen R and D, Contracted Research, Self, Neurocrine, Contracted Research, Self, Janssen Scientific Affairs, Consultant, Self, Supernus, Contracted Research, Self, Boehringer Ingelheim, Consultant, Self, Elsevier, Honoraria, Self, Worldwide Clinical Trials, Honoraria, Self, Vicore Pharma, Honoraria, Self, IQVIA, Honoraria, Self, Psych Congress, Honoraria, Self, Medscape/WebMD, Honoraria, Self, Physician's Education Resource, Honoraria, Self, Clinical Care Options, Honoraria, Self, H.C. Wainwright and Co., Honoraria, Self.

P151. FROM CLINICIAN TRAINING TO SCALE-SPECIFIC CONVENTIONS: A NOVEL APPROACH FOR LLM-BASED RATER AND DATA QUALITY CONTROL

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Background: Development of semi-structured interviews, such as the SIGMA (MADRS) and SIGH-A (HAM-A), offered anchored severity ratings, along with standardized questions and scoring conventions to subjective CNS scales. Underlying the high inter-rater reliability of these structured interviews was the assumption that the raters administering the scales had extensive clinical training that included interview skills and the ability to interpret symptom severity across multiple therapeutic areas.

A common practice for rater and data quality control monitoring in today's CNS trials includes recording of primary outcome interviews on scales such as the MADRS, HAM-D, HAM-A and PANSS, with a second independent rater reviewing the recording and creating a second set of scores.

Use of Artificial Intelligence (AI) and Large Language Models (LLMs) for rater and data quality control is emerging and now a standard component of EMA Wellness' eCOA and data surveillance platforms. Trained LLM models generate a standardized second set of scores for CNS scales in studies, which can be used to identify potential scoring or administration issues. To date, most LLM models have been developed with inputs and prompts on specific ratings scales, such as the MADRS and HAM-A, and challenges interpreting nuanced symptoms like a trained human clinician exist.

This analysis focuses on the development of an LLM that is similar to an experienced clinician that can interpret reported symptoms across multiple therapeutic areas, and is then trained to apply those abilities on the specific scoring and administration conventions of individual scales.

Methods: A private GPT-4o LLM hosted on a Microsoft Azure server was provided inputs of an interview skills training and placebo response training that raters in multiple therapeutic areas may complete. Additional custom inputs by experienced clinicians on evaluating severity, frequency and impact in clinical interviews was provided to the model.

The model was then provided the inputs of a SIGH-A rating guide and a HAM-A rater training that featured the SIGH-A. 50 audio recorded HAM-A interviews from multiple late phase GAD studies were transcribed.

The same model was also provided the inputs of a SIGMA rating guide and a MADRS rater training that features the SIGMA. 50 audio recorded MADRS interviews from multiple late phase studies were transcribed.

The model scored each item from both the HAM-A and MADRS interviews based on the transcriptions, and included a detailed scoring rationale for each item. These scores were then compared to the interviewing rater's scores.

Results: The interview rater scores and LLM model scores were paired and analyzed for 50 HAM-A interviews and 50 MADRS interviews. The ICC for all items of the 50 HAM-A interviews was 0.79, and the mean total score the rater was 23.1, LLM 22.8. 97% of all items had a discrepancy between ratings of one point or less. The ICC for all items of the 50 MADRS interviews was .78, and the mean total score of the rater was 14.2, LLM 25.8. 86% of all items had a variance of one point or less.

Conclusions: The effectiveness and scalability of validated LLM models for rater and data quality control in CNS studies provides the opportunity to monitor clinical outcomes like the MADRS and HAM-A by flagging potential rater drift, interview quality and anomalous scores in real-time. The issues identified by these models can then be escalated to expert human clinicians. The clinical utility of an LLM that is trained first as "clinician analogues" before then training on scale-specific conventions should enhance the LLMs ability to interpret nuanced symptoms across therapeutic areas and scales. As a trained LLM receives inputs from multiple scales, trainings and therapeutic areas, the clinical expertise of the model should continue to grow.

Keywords: Artificial Intelligence, Analysis, NeuroScience, Clinical NeuroScience, Pharmacology, generalized anxiety disorder, MADRS, HAM-A

Disclosure: EMA Wellness, Founder, Self.

P152. HARNESSING THE ENDOCANNABINOID SYSTEM TO ENHANCE PROLONGED EXPOSURE THERAPY: PRELIMINARY EVIDENCE FROM RANDOMIZED TRIAL OF D9-THC IN PTSD

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Background: Posttraumatic stress disorder (PTSD) is characterized by intrusive memories, avoidance, negative cognitions, and hyperarousal, leading to significant impairment. Prolonged exposure (PE) is the gold-standard treatment, yet many patients remain symptomatic or discontinue. The endocannabinoid system, which regulates stress and extinction learning, is a promising target. Preclinical work suggests cannabinoids may facilitate extinction and improve affect regulation. We tested whether adjunctive D9-tetrahydrocannabinol (THC) enhances PE outcomes in PTSD.

Methods: Twenty-three adults meeting DSM-5 PTSD criteria were randomized (double-blind) to 7.5 mg oral THC (n = 11) or placebo (n = 12) administered during four of ten PE sessions. Clinical outcomes were assessed with the Clinician-Administered PTSD Scale (CAPS-5, primary), and PTSD Checklist (PCL_5), alongside secondary measures of depression (HAM-D, BDI-II), anxiety (HAM-A), cognitive avoidance (CAQ), and sleep quality (PSQI). Linear mixed-effects models examined trajectories of change. Sex was included as a covariate. This study was not powered for definitive efficacy but powered to detect medium effect sizes. Trial Registration: NCT04080427. Adverse events were systematically tracked. No serious adverse events occurred.

Results: Both groups showed robust improvements in PTSD symptoms (CAPS-5, mean $\Delta = -26.77$ (SD = 15.12), $d = -1.78$, $p < 0.0001$; PCL-5, mean $\Delta = -31.94$ (SD = 15.41), $d = -2.13$,

$p < 0.0001$), depression (HAM-D, mean $\Delta = -5.02$ (SD = 6.08), $d = -0.97$, $p = 0.003$; BDI-II, mean $\Delta = -12.00$ (SD = 12.77), $d = -0.94$, $p = 0.03$), cognitive avoidance (CAQ, mean $\Delta = -34.05$ (SD = 19.9), $d = -1.71$, $p = 0.001$), maladaptive cognitions (PTCI, mean $\Delta = -49.86$ (SD = 40.65), $d = -1.23$, $p < 0.0001$), and sleep quality (PSQI, mean $\Delta = -3.86$ (SD = 2.73), $d = -1.44$, $p = 0.0004$). Anxiety was not significantly affected (HAM-A, mean $\Delta = -2.86$ (SD = 7.67), $d = -0.45$, $p = 0.12$). At 3-month follow-up, THC produced greater reductions than placebo on CAPS-5 ($\Delta\Delta = -3.55$ points (95% CI: -10.03 to -1.28), $d = -1.23$, $p = 0.012$) further, HAM-D score was significantly reduced with THC ($\Delta\Delta = -6.36$ points (95% CI: -8.90 to -3.82), $d = -1.48$, $p = 0.021$) but not for PCL ($\Delta\Delta = -3.67$ points (95% CI: -8.11 to 0.78), $d = -0.47$, $p = 0.036$), however these differences were not significant between groups ($p = 0.53$). There was a single effect of Sex on BDI-II ($p = .03$).

Conclusions: This preliminary randomized trial suggests THC may augment PE therapy for PTSD, particularly for clinician-rated PTSD severity. This study was not powered for definitive efficacy but was powered to detect medium effect sizes, consistent with the observed effects. These findings support further investigation of cannabinoid-assisted psychotherapy in larger, dose-ranging, studies

Keywords: THC, PTSD, Prolonged Exposure Therapy

Disclosure: Nothing to disclose.

P153. VIRTUAL RESIDENTIAL ALCOHOL USE DISORDER TREATMENT USING BREATHALYZERS WITH WIRELESS CONNECTIVITY AND FACIAL RECOGNITION TO MONITOR ABSTINENCE

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Background: Despite recent increases in alcohol-related mortality, access to alcohol use disorder (AUD) treatment remains challenging. Accessing publicly-funded residential treatment programs is particularly difficult due to long wait times. Virtual programs that incorporate technology to enhance treatment have the capacity to improve access to care, including providing care to regions that lack specialized treatment facilities. We aimed to develop an entirely virtual treatment program for AUD which would replicate the structure and abstinence-monitoring of a residential treatment program without the need for a brick-and-mortar facility. We employed take-home breathalyzers with data connectivity and facial recognition technology to monitor abstinence. Here, we present data on the feasibility and efficacy of this program as well as data on adherence and satisfaction with the breathalyzer component of the program.

Methods: In this clinical trial, 56 participants (28 male, 28 female) with moderate to severe AUD were enrolled and started the treatment program. During the 4-week program, patients were treated with three manualized therapies: a cognitive behavioral therapy for AUD group (12 sessions, delivered 3 times per week), a dialectical behavior therapy skills groups (12 sessions, delivered 3 times per week), and a health and wellness group (8 sessions, delivered twice per week). Each day, participants were instructed to complete breathalyzer readings at four scheduled intervals. Outcomes included retention in treatment, change in drinking during treatment (measured with weekly Timeline Followback interviews), satisfaction with the overall program (measured using the Client Satisfaction Questionnaire-8 [CSQ-8]), and adherence to and satisfaction with the breathalyzer component of the program. Measures of central tendency were

reported as mean and standard deviation (SD) or median and interquartile range (IQR). A paired t-test was used to compare drinking at the start and end of the program.

Results: 91.1% of patients completed the treatment program. Alcohol intake was reduced from a mean of 3.5 (SD = 3.5) standard drinks per day at the start of the program to 0.8 (SD = 1.6) standard drinks per day in the last week of treatment ($t(51) = 5.0$, $p < 0.001$, Cohen's $d = 0.70$). The median percentage of completed breathalyzer readings at the scheduled time interval was 96.0% (IQR = 88.0 to 99.0). The median number of positive breathalyzer tests throughout the program was 1.5 (IQR = 0.0 to 7.5) out of 112 possible prompts. Client satisfaction with the treatment program was high, with a median CSQ-8 score of 30.0 (IQR = 28.0 to 32.0) out of 32. Over 90% of participants agreed or strongly agreed that they were satisfied with the breathalyzer component of the study and that breathalyzer usage helped them attain their treatment goals.

Conclusions: These results demonstrate the feasibility of this novel treatment program and indicate good uptake of the breathalyzer component of the program.

Keywords: Alcohol Use Disorder, Telemedicine, Technology

Disclosure: Nothing to disclose.

P154. EFFECT OF ADJUNCTIVE N-ACETYLCYSTEINE VS PLACEBO FOR CLINICAL AND FUNCTIONAL OUTCOMES IN ADOLESCENTS WITH FIRST-EPISEDE PSYCHOSIS: RESULTS OF A DOUBLE-BLIND RANDOMIZED TRIAL

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Background: First-episode psychosis (FEP) in adolescents presents a complex clinical profile, often involving psychotic, affective, and functional impairments that extend beyond symptoms responsive to standard antipsychotic treatments. Given the partial efficacy and side effects of antipsychotics, there is a pressing need to explore adjunctive medications that target underlying neuropathological mechanisms. N-Acetylcysteine (NAC), an antioxidant and glutathione precursor, has shown promise in adult schizophrenia through modulation of oxidative stress and glutamatergic pathways, with meta-analyses indicating modest improvements in negative symptoms and general psychopathology when used in augmentation to antipsychotics (PMID: 37852631). Evidence in adolescents, however, remains limited highlighting the need for specifically designed trials in this population to evaluate NAC's potential as a safe, adjunctive treatment.

Methods: This double-blind, multicenter, phase 3 trial randomized male and female adolescents (aged 12 to < 18 years) experiencing early-onset first psychotic episodes to receive oral N-Acetylcysteine (NAC) or matched placebo (1:1) twice daily for 48 weeks (EudraCT Number: 2012-005435-87). NAC doses ranged from 1200 to 3000 mg/day. All participants received standard antipsychotic treatment alongside study medication. Eligible patients met DSM-IV criteria for psychotic disorders (F20 or F30), confirmed via K-SADS interview.

The primary outcome was change in frontal gray matter volume at 48 weeks. Here, we report on the clinical secondary outcomes, that included between-group differences in psychopathology and function—Positive and Negative Syndrome Scale (PANSS) total, positive, negative, and general subscales, Young Mania Rating Scale (YMRS), Hamilton Depression Rating Scale (HAM-D-17), Children's Global Assessment Scale (C-GAS), and Social Functioning Assessment Scale (SOFAS)—assessed at 24 weeks, 48 weeks (end of treatment), and 4 weeks post-treatment. Sample size was powered

based on our prior cohort data on gray matter loss (PMID: 22213785), hypothesizing NAC would halve loss versus placebo. Eighty patients were required for 80% power at $\alpha = 0.05$, increasing the expected recruitment number to account for attrition.

Analyses used intention-to-treat mixed models for repeated measures, adjusting for baseline variables of interest, sex, age, race/ethnicity, socioeconomic status (measured via Hollingshead and Redlich scale), center, and Body Mass Index (BMI). False discovery rate correction addressed multiple comparisons.

Results: A total of 96 adolescents were randomized to adjunctive NAC ($n = 47$) or placebo ($n = 49$), with groups showing well-matched sociodemographic and baseline clinical characteristics. Mean (SD) age was 15.57 (1.35) years in NAC and 15.60 (1.33) years in placebo; females accounted for 59% and 57% of each group respectively. Socioeconomic status mean (SD) was 42.62 (19.79) for NAC and 38.42 (17.81) for placebo. Distribution by race/ethnicity was European 64% vs 62%, South/Central American 27% vs 28%, and Other 9% vs 10% in NAC and placebo, respectively. Baseline clinical scores were not significantly different between groups (all $p > 0.05$): PANSS positive 15.40 (6.41) vs 15.98 (6.12), PANSS negative 19.70 (9.23) vs 21.71 (7.43), PANSS general 33.21 (11.12) vs 37.20 (12.51), and PANSS total 67.12 (19.68) vs 74.89 (21.79). YMRS was 7.52 (9.46) vs 6.89 (7.02), HAM-D-17 was 8.12 (6.02) vs 9.16 (7.73), C-GAS was 44.30 (12.80) vs 43.28 (12.16), SOFAS was 45.77 (14.75) vs 45.45 (14.11), BMI was 22.20 (4.19) vs 22.95 (4.64), and cumulative chlorpromazine equivalents were 19,287.18 (28,285.94) vs 17,661.01 (21,168.18) for NAC and placebo, respectively.

Mixed model analyses for repeated measures revealed that, at 48 weeks (end of treatment), the placebo group had significantly lower adjusted mean scores than NAC for total PANSS (mean difference 7.42; 95% CI, 0.97–13.86; $p = 0.02$) and PANSS general (mean difference 3.97; 95% CI, 0.48–7.45; $p = 0.03$). These differences, however, were not statistically significant after false discovery rate correction (both $p = 0.47$). No significant between-group differences were observed for PANSS positive, PANSS negative, YMRS, HAM-D-17, C-GAS, or SOFAS at 48 weeks. No significant between-group differences were found in any clinical or functional outcomes at 24 weeks or 4 weeks post-treatment.

Conclusions: Adjunctive NAC did not demonstrate significant clinical or functional benefits over placebo after 48 weeks in adolescents with first-episode psychosis. Despite NAC's theoretical and adult data rationale, further research is warranted to assess its role in early psychosis, perhaps targeting specific symptom domains or considering biomarker-guided treatment.

Keywords: first-episode psychosis, N-acetylcysteine, Adolescent

Disclosure: Rovi, Honoraria, Self, Teva, Honoraria, Self, Outsuka, Honoraria, Self, Angelini, Honoraria, Self, Lundbeck, Honoraria, Self, Janssen, Honoraria, Self.

P155. SAFETY OF CO-ADMINISTERED CANNABIDIOL (CBD) AND ALCOHOL IN ACUTE LABORATORY AND CHRONIC SETTINGS: A PHASE I STUDY

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Background: Cannabidiol (CBD) and alcohol co-use is growing increasingly common, yet the safety of this behavior, including hepatic effects, is not well-understood. This Phase I study aimed to assess the safety and tolerability of CBD and alcohol co-administration among moderate alcohol consumers in acute and chronic outpatient settings. The CBD doses were determined as best reflecting the amounts that typical consumers use while

remaining within the maximum amount that is recommended for safe use among healthy adults.

Methods: The sample consisted of healthy adults ages 21–65 with moderate alcohol use ($n = 19$). The sample predominantly identified as white, non-Hispanic males. Average number of drinks per week at baseline was 11.1 with a standard deviation of 11.3 drinks. Participants completed three acute dosing sessions where they randomly received placebo, 50 mg CBD, or 100 mg CBD along with alcohol dosed to achieve peak blood alcohol concentrations (BAC) of 0.06 g/100 mL. BAC. Measures of craving, sedating and stimulating effects, and impairment were collected from participants throughout each session. Participants then consumed 50 mg CBD twice daily for four weeks while reporting alcohol use, craving, and adverse effects via a smartphone app. Metabolic labs and CBD content were measured from blood samples collected at the end of each experimental appointment and at four-week follow-up. Data were analyzed using linear mixed effect models with timepoint and CBD dose as factors. Laboratory data were compared using paired t-tests and a false discovery rate correction. Adverse events and drinking outcomes from the chronic dosing period were collected and descriptively tabulated. The protocol was approved by the WIRB-Copernicus Group (WCG) Institutional Review Board (Study number:1328975). The study was registered on Clinicaltrials.gov under record number NCT05317507.

Results: Physiologic and subjective responses from experimental sessions showed prototypic alcohol effects (e.g., stimulatory effects on the ascending limb of BAL curve, $p < 0.001$). No significant differences in BAL timecourse or peak were observed with CBD co-administration at any dose ($p = 0.12$). CBD did not alter peak stimulatory or sedentary effects of alcohol ($p > 0.18$). Limited adverse effects were reported; CBD had no consistent effect on number of outpatient drinks consumed. No significant changes in chemistries, including LFTs, were observed after four weeks of CBD administration apart from a non-clinically significant decrease in protein, (0.23 g/dL decrease, $p = 0.006$).

Conclusions: CBD co-administration with alcohol was not associated with significant subjective or physiologic effects among moderate alcohol consumers in acute laboratory and naturalistic outpatient settings. Results support the acute and chronic safety and tolerability of CBD-alcohol co-use in further studies. The safety of CBD use in patients with more severe alcohol use should be examined in future studies.

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Competing Interests: JS has received research related funding from Canopy Growth Corporation and DynamiCare Health and consulting fees from Realm of Care Foundation and Merck Corporation in the past three years. EW has received research support from Canopy Growth Corporation for clinical research projects unrelated to this study. AH is a former employee of Canopy Growth Corporation. ENP received stock options as a former employee of Canopy Growth Corporation during execution of study. MBM is an employee of Charlotte's Web, a member of the Board of Directors at DeFloria, and a former employee of Canopy Growth Corporation.

Keywords: Cannabidiol, Alcohol consumption, Phase 1

Disclosure: Nothing to disclose.

P156. NEGATIVE SYMPTOMS AND SEDENTARY BEHAVIOR IN OLDER PARTICIPANTS WITH SCHIZOPHRENIA: BASELINE FEATURES AND EFFECTS OF 12 MONTHS OF TREATMENT WITH XANOMELINE/TROSPIMUM

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Background: Previous research suggested that negative symptoms may worsen with age in schizophrenia. Elements of negative symptoms, such as avolition, anergia, and amotivation, are defined by in part by reduced physical activity. Thus, we examined age-related differences in negative symptoms measured by active and passive digital phenotyping in a 12-month open-label trial with xanomeline/trospium chloride (X/T). Assessments included 3 times per day momentary reports of everyday activities and concurrent measures of steps captured with actigraphy. We also report month x month treatment-related changes across older and younger age groups in an effort to understand both age-related baseline performance and changes with treatment

Methods: Participants included 341 male and female outpatients with schizophrenia (212 > age 45 years). All completed remote ecological momentary assessment (EMA) surveys 3 times per day, 7 days per week, one week per month for up to 12 months. On EMA survey days, participants wore a Fitbit™ actigraph that recorded their steps. EMAs queried location, social context, positive affect (PA), and activities. For activity analyses, EMA-measured activities were characterized as recumbent (resting/sleeping), seated, standing, and moving. We examined baseline age differences and changes over time.

Results: Participants answered 40,464 EMA surveys and provided 7957 participant-days of actigraphy. There were significant treatment month-related reductions with X/T treatment in EMA-reported recumbent and seated activities, as well as increases in standing and movement both at home and away, all $X^2 > 54.5$, all $p < 0.001$. Steps significantly increased over the 12 months, $X^2 = 22.84$, $p = 0.019$. Higher momentary PA was associated with more daily steps, $X^2 = 91.83$, $p < 0.001$, and a significant interaction of month x PA, $X^2 = 144.99$, $p = 0.009$, suggested an increasing impact of PA on step counts over time. Increased step counts predicted reductions in recumbent and seated activities, with very large associations, $X^2 = 1874$, $p < 0.001$ and $X^2 = 1503$, $p < 0.001$, respectively. At baseline, older participants were home more frequently ($p < 0.001$) and had significantly lower step counts, ($p < 0.001$), engaging in more recumbent and seated activities (both $p < 0.001$) and fewer standing and moving activities (both $p < 0.01$). Their baseline PA scores were higher ($p < 0.001$). All treatment effects were significant in older participants when examined alone and there were no interactions of age group x month, suggesting equivalent improvements over time despite greater baseline sedentary activities.

Conclusions: Although older participants with schizophrenia started the treatment study with increased sedentary behavior and other negative symptoms compared with younger individuals, the treatment-related benefits were statistically significant and of similar magnitude across age. Previously reported effects of changes in PA on negative symptoms were replicated in both older and younger participants. These data suggest that there are age-related differences in physical activities related to negative symptoms in schizophrenia but that these features can change with treatment, possibly leading to improvements in health outcomes.

Keywords: Negative Symptoms, Sedentary Behavior, xanomeline, aging with schizophrenia

Disclosure: BMS, Consultant, Self, Alkermes, Consultant, Self, Kynexis, Consultant, Self, Minerva Neurosciences, Consultant, Self, Neurocrine Biosciences, Consultant, Self, i-Function, Founder, Self, EMA Wellness, Founder, Self, WCG endpoint solutions, Royalties, Self.

P157. A PILOT RANDOMIZED CONTROLLED TRIAL OF NITROUS OXIDE INHALATION IN UNMEDICATED ADULTS WITH OCD: PROOF OF CONCEPT

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Background: The inhaled anesthetic nitrous oxide (N2O) exhibits ketamine-like pharmacodynamic and antidepressant effects but has not been studied in OCD. We report preliminary results through July 2025 from an ongoing IRB-approved, randomized placebo-controlled pilot study of N2O gas inhalation in unmedicated adults with OCD (NCT03826693).

Methods: N = 20 adults (mean age 36, range 21–65, 50% female) with moderate to severe DSM-5 OCD were randomized 2:1 to a single-blind 1-hour inhalation of 50% N2O/50% oxygen vs air/oxygen placebo. A blinded independent rater assessed Y-BOCS at baseline and post-inhalation days 1, 3, 7, and 14. Secondary outcomes included measures of insight, depression, and acute dissociative symptoms. Exploratory biomarkers assessed pre- and post-inhalation included serum prolactin and an EEG event related potential (ERP) indexing stimulus incongruity during an arrow flanker task. Outcomes were assessed with linear mixed effects modeling.

Results: N2O was well tolerated with no significant dissociative effects. A robust but non intervention-specific total Y-BOCS response was evident at post-inhalation Day 1, with gradually increasing separation between active and placebo arms over the two-week follow up period. A statistically significant, moderate to large effect of N2O vs placebo on total Y-BOCS was observed at Day 14 ($\beta \pm SE -4.6 \pm 2.28$, Hedges' $g = -0.74$, $p = 0.05$). A large, significant effect of N2O vs placebo on Y-BOCS obsessions was observed at Day 7 ($\beta \pm SE -2.52 \pm 1.20$, $g = -0.80$, $p = 0.04$) and at Day 14 ($\beta \pm SE -2.88 \pm 1.20$, $g = -0.92$, $p = 0.02$), but no significant effect was observed on Y-BOCS compulsions. N2O vs placebo increased a midline-central (Cz) ERP biomarker indexing stimulus incongruity at one hour post inhalation ($\beta \pm SE -2.52 \mu V \pm 0.70$, $p = 0.003$), and acute change in this ERP marker correlated with Y-BOCS obsessions change at Day 14 ($r = 0.57$, $p = 0.01$).

Conclusions: N2O alleviated OCD symptoms up to two weeks after a 1-h inhalation and altered an ERP measure of stimulus incongruity processing. The promising preliminary findings of this pilot study provide first evidence of N2O's impact on both clinical symptoms of OCD and an EEG/cognitive control biomarker and warrant replication within a larger sample.

Keywords: Obsessive-Compulsive Disorder (OCD), Nitrous Oxide, Randomized Clinical Trial, Flanker Task, EEG biomarkers

Disclosure: Nothing to disclose.

P158. BASELINE LEVELS OF INFLAMMATION AS A MODERATOR OF ALCOHOL CRAVING AND DRINKING IN A CLINICAL TRIAL OF A NEUROIMMUNE MODULATOR FOR ALCOHOL USE DISORDER

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Background: Individuals with alcohol use disorder (AUD) often exhibit heightened levels of systemic inflammation, which is hypothesized to contribute to continued alcohol use. Thus, the inflammatory system may serve as a promising biological target for novel medications for AUD. This study is a secondary analysis of a recently completed 12-week randomized controlled trial testing ibudilast, a phosphodiesterase inhibitor with anti-

inflammatory properties, as a treatment for AUD (ClinicalTrials.gov Identifier: NCT03594435). Primary findings from the trial did not reveal significant differences between ibudilast and placebo on the primary drinking outcome, percent heavy drinking days (PHDD). However, both groups significantly reduced their drinking across the trial. Ibudilast was associated with a greater decrease in alcohol craving across the trial compared to placebo. The current study tested whether baseline levels of peripheral inflammation moderated the effects of ibudilast on alcohol craving and drinking outcomes. Given its proposed anti-inflammatory effects, we hypothesized that ibudilast would be more effective at reducing craving and drinking among individuals with elevated levels of baseline inflammation.

Methods: Ninety-nine treatment-seeking individuals with moderate-to-severe AUD were randomized to receive ibudilast (50 mg/twice daily; $n = 50$ [31M/19F]) or placebo ($n = 49$ [29M/20F]) for 12 weeks. Participants provided blood samples at baseline, prior to medication titration, to measure circulating levels of C-reactive protein (CRP) and peripheral cytokines. Concentrations were log-transformed to normalize distributions, and Pearson correlations were calculated to examine associations between the markers. CRP, interleukin-6 (IL-6), and tumor necrosis factor alpha (TNF- α) showed the strongest positive correlations (Pearson $r > 0.30$) and were combined into an inflammatory composite score by averaging the Z-scores of their logged concentrations. Alcohol craving was measured using the Penn Alcohol Craving Scale (PACS) at baseline and at weeks 4, 8, and 12 of the trial. Alcohol use (PHDD) was assessed at baseline and biweekly via the Timeline FollowBack. Mixed-effects models in SAS 9.4 tested the three-way interaction between medication group, baseline inflammation levels, and time on alcohol craving and drinking trajectories, with the inflammation composite modeled as a continuous predictor. Craving was analyzed using linear mixed models, while alcohol use was modeled with two-phase piecewise linear mixed models to capture distinct patterns observed across time. Random intercepts and slopes were included in the models. Sex, medication compliance, cigarette and cannabis use status, and changes in drinking (craving model only) were included as covariates.

Results: There was a significant 3-way interaction between medication group, baseline inflammation levels, and time ($\beta = 0.59$, $SE = 0.27$, $p = .03$) on alcohol craving, such that higher levels of baseline inflammation were associated with greater reductions in craving over the trial for participants receiving ibudilast compared to placebo. In contrast, baseline inflammation did not moderate heavy drinking trajectories by medication group for either phase of the trial (Phase 1: $\beta = -0.14$, $SE = 0.12$, $p = .25$; Phase 2: $\beta = 0.17$, $SE = 0.13$, $p = 0.19$).

Conclusions: These findings indicate that ibudilast, a neuroimmune modulator, may be more effective for individuals with high levels of baseline inflammation in reducing craving for alcohol but not drinking. This may reflect greater capacity for reductions in inflammation among individuals with elevated baseline inflammation compared to those with low baseline inflammation, who may exhibit a floor effect. The results are consistent with ibudilast's proposed anti-inflammatory mechanism. Although alcohol craving and consumption are interrelated, the lack of medication effects on drinking may reflect behavioral improvements related to placebo effects and the motivation associated with seeking treatment, as both groups dramatically decreased drinking over the trial. In contrast, craving assessments capture subjective experiences that may be less influenced by expectancy effects. Together, these results suggest that baseline inflammation may help identify individuals most likely to benefit from anti-inflammatory medications for AUD and highlight the importance of integrating biomarkers into clinical trials to inform targeted treatment strategies.

Keywords: randomized clinical trial, Alcohol Use Disorder - Treatment, inflammation, pharmacotherapy

Disclosure: Nothing to disclose.

P159. A FIRST-IN-HUMAN STUDY OF NS-136, A NOVEL M4 MUSCARINIC RECEPTOR POSITIVE ALLOSTERIC MODULATOR: SAFETY, TOLERABILITY, AND PHARMACOKINETICS IN HEALTHY ADULT AND ELDERLY SUBJECTS IN AUSTRALIA AND CHINA (NS136HV101)

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Background: NS-136 selectively targets the M4 muscarinic receptor through positive allosteric modulation. It acts on presynaptic muscarinic autoreceptors, which indirectly regulate dopamine release by reducing acetylcholine release. This novel mechanism is promising for the treatment of symptoms in schizophrenia without blocking the postsynaptic dopamine D2 receptors. To better understand the ethnic differences and prepare for the clinical development covering broad patient population, we conducted an FIH study in both Australia and China to assess the safety, tolerability, and pharmacokinetics of NS-136 both in healthy adult and elderly subjects.

Methods: Australia Part: The single ascending dose (SAD) phase enrolled 40 healthy volunteers, randomized in a 6:2 ratio to receive NS-136 (5, 15, 30, 40, or 55 mg per day) or placebo. The multiple ascending dose (MAD) phase included 16 healthy volunteers, randomized in a 6:2 ratio to receive NS-136 (20 or 40mg) or placebo once daily for 14 days.

China Part: A crossover cohort of 12 participants assessed food effects (FE) using a 50 mg dose as a bridging dose from Australia to China. Given the favorable safety profile, the SAD phase continued with doses of 65, 80, 100, and 120 mg, while the MAD phase included doses of 50, 65, 80, 100 and 120 mg per day. Starting from the 80 mg/day cohort, the SAD and MAD dose escalations were combined, which was with a single dose on day 1, followed by safety observations for 3 days and the multiple dosing occurred from day 4 to day 17 (14 days). In addition, a separate MAD phase comprising 32 elderly healthy volunteers aged 65 to 85 years, randomized in a 6:2 ratio to receive NS-136 (50, 70, 90 or 120 mg) or placebo was included to evaluate the safety, tolerability, and pharmacokinetics of NS-136 in elderly population. Serial plasma pharmacokinetic (PK) samples were collected, and safety assessments included monitoring for adverse events (AEs), laboratory tests, electrocardiograms (ECGs), and physical examinations.

Results: All reported TEAEs were mild and self-resolving. No moderate or severe TEAEs or clinically significant laboratory or ECG abnormalities were observed. A transient increase in heart rate was observed following dosing and recovered. NS-136 exhibited a terminal half-life of 10 to 16 h when administered above 50 mg QD after multiple dosing, supporting once-daily administration. Pharmacokinetic exposure increased subproportionally with dose in both the SAD and MAD cohorts. Under fed conditions, there were slight delays in C_{max}, although overall exposure was unaffected. Furthermore, no significant differences in exposure were observed between the Australian and Chinese populations. Besides, the safety and pharmacokinetic profiles in elderly participants were favorable and showed no significant differences compared to those in younger adults.

Conclusions: In total (n = 148) healthy volunteers enrolled in NS-136 phase 1 studies, including 56 adults in Australia, 60 adults and 32 elderly in China, NS-136 demonstrated favorable safety

and tolerability up to 120 mg, with PK properties suitable for once-daily administration without titration. No major ethnic differences between Australian and Chinese population observed, as well as elderly population, which supports multi-regional studies in phase 2 or beyond. A Phase 2a proof-of-concept study was initiated in patients with schizophrenia. Furthermore, a separate Phase 2a trial in elderly subjects with agitation in Alzheimer's disease is under planning.

Keywords: Schizophrenia (SCZ), M4 muscarinic acetylcholine receptor (mAChR), positive allosteric modulators, Clinical trial, phase 1 study

Disclosure: NeuShen Therapeutics, Employee, Self, Shinesta Medicine, Employee, Spouse/Partner.

P160. EXPLORING THE PHARMACOKINETIC AND PHYSIOLOGICAL INTERACTIONS BETWEEN THC AND CBD: A PLACEBO-CONTROLLED HUMAN LABORATORY STUDY

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Background: Cannabis legalization led to increased usage, perceived safety, and potency of Δ9-tetrahydrocannabinol (THC, main intoxicating component of cannabis). There is growing evidence that cannabinoids are an effective treatment for several health conditions; but increased usage and potency are also associated with more adverse effects (psychiatric effects, cognitive impairment, cardiovascular effects, and abuse liability). Previous research suggests that CBD, a non-intoxicating cannabis component, may reduce the adverse effects of THC. However, other studies have found no CBD modulatory effects or that oral CBD administration actually leads to increased THC blood concentrations and enhanced effects. To further clarify the potential interactions between THC and CBD, this study investigated THC and CBD effects using the most common route of administration (smoking) on endpoints consistently used to determine safety and adverse effects: cognition, cardiovascular effects, subjective drug effects, and abuse liability. Metabolism of THC and CBD was examined to determine if effects are due to a pharmacokinetic interaction between the two cannabinoids.

Methods: Healthy adults (21–55 years) with infrequent cannabis usage were recruited for this placebo-controlled, within-subject study. Exclusion criteria included use of prescription medication, diagnosis of substance use or major psychiatric disorder, and pregnancy/lactation. Participants were instructed to remain cannabis abstinent throughout the study. Participants smoked 500 mg cannabis plant material with either placebo (PBO, 0 mg), THC (20 mg), CBD (20 mg), or combined THC + CBD (20 mg each) in a randomized order across four sessions. The following outcomes were assessed prior to and across several timepoints after exposure: (1) pharmacokinetics (blood THC, CBD, and their metabolites concentrations); physiology (heart rate); cognition (assessed via DRUID app [Impairment Sciences, Inc.]); and subjective drug effects (assessed via Mood and Physical Symptoms Visual Analog Scales [MPS-VAS] and Cannabis Rating Form [CRF]). Repeated measures ANOVA tested the effect of cannabis composition (PBO, THC, CBD, THC + CBD) on all outcome measures across time (p < 0.05).

Results: Twelve healthy adults (28.3 ± 7.7 years old, 2 female) who reported infrequent cannabis use (<1 day per week, ~3x per

month) completed the study. Peak blood THC concentrations (timepoint [T]10) were elevated in THC and THC + CBD sessions ($p < 0.05$), and peak blood CBD concentrations were elevated in CBD sessions ($p < 0.05$) compared to PBO. CBD did not elevate blood THC concentrations; and THC did not elevate blood CBD concentrations. THC (THC and THC + CBD) resulted in elevated heart rate (T10 and T30, $p < 0.015$), increased intoxication ("I feel high": T30 and T60, $p < 0.05$), impaired cognition (DRUID scores: T10 and T30, $p < 0.05$), and greater subjective drug effects related to feeling impaired and drug strength ("I feel impaired": T90 and T120, $p < 0.05$; "strength of effect": $p < 0.05$), compared to CBD and PBO. Some THC effects were mitigated when THC + CBD were co-administered, including reduced duration of intoxication and elimination of subjective cognitive impairments. Specifically, compared to PBO and CBD, THC alone resulted in effects on intoxication ("I feel high": T10, T90, T120, $p < 0.05$; "I feel impaired": T10, T30, T180-T360, $p < 0.05$) and subjective cognition ("I feel forgetful", $p < 0.003$) that were not seen with THC + CBD. CBD alone did not differ from PBO.

Conclusions: This study replicates previous research showing the effects of THC on physiology, cognition, intoxication, and subjective drug effects. Interestingly, co-administration of THC + CBD mitigated some of the effects of THC on intoxication and endpoints related to impairment. This study provides important evidence for the harm reduction potential of CBD. Future research will examine whether higher CBD doses can better attenuate THC's adverse effects. This research was funded by: California Highway Patrol (CHP), NIDA DSP for Cannabis, and Semel Charitable Foundation to ZD Cooper. Canadian Institute of Health Research (CIHR) postdoctoral fellowship to SL Baglot and S Lake.

Keywords: cannabis, delta9-tetrahydrocannabinol (THC), cannabidiol (CBD), adverse effects, harm reduction

Disclosure: Nothing to disclose.

P161. INCREASING NAD⁺ LEVELS IS ASSOCIATED WITH IMPROVED SLEEP QUALITY IN LONG-COVID

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Background: Sleep disturbances are common in long-COVID, a syndrome characterized by persistent and often debilitating symptoms lasting for months or even years post SARS-CoV-2 infection. SARS-CoV-2 infection has been shown to dysregulate nicotinamide adenine dinucleotide (NAD⁺)-dependent pathways, a critical coenzyme involved in cellular metabolism and immune regulation. Preclinical studies suggest that NAD⁺ supplementation can impact sleep. In this study, we examined whether increasing NAD⁺ levels using nicotinamide riboside (NR), a NAD⁺ precursor, could improve sleep quality in adults with long-COVID.

Methods: Fifty-eight community-dwelling participants with long-COVID participated in a 24-week, double-blind, placebo-controlled clinical trial with a placebo lead-in phase (NCT04809974) at the Massachusetts General Hospital. Participants were infected with SARS-CoV-2 at least two months prior to enrollment and were never hospitalized. Eligible participants completed a two-week PBO lead-in period. Baseline measures were then collected, and participants were randomized 2:1, based on their sequence, to receive either 2000 mg of oral Niagen NR (64%) daily or placebo (PBO) (34%) for 10 weeks. After 10 weeks, people in the PBO group switched to the NR phase (PBO-NR) for

10 weeks, while those in the NR group stayed in the NR phase (NR-NR). Of the total, 25 and 18 participants in the NR-NR group remained in the study at 10 and 20 weeks after baseline, respectively, and 18 in the PBO-NR group. The Pittsburgh Sleep Quality Index was administered as a secondary outcome measure to assess self-reported sleep quality at baseline, 5, 10, 15, and 20 weeks. NAD⁺ levels were measured in whole blood at 5, 10, 15, and 20 weeks. We performed intention-to-treat analysis and mixed-effects model repeated measures to compare groups. We also did a sequential block design as a post-hoc analysis. This provided the opportunity to examine the effects of NR using a larger sample size since all participants ultimately received 10 weeks of NR. We performed linear regression analyses of PSQI change scores and NAD⁺ levels measured after 10 weeks. Models included age, sex, education, and time since acute infection.

Results: On average, participants in the NR-NR group were 48.5 years old and mostly female (83%), and in the PBO-NR group 41.7 years old and mostly female (57%). The average compliance rates were above 96% in both groups. In the NR-NR group, NAD⁺ levels increased by an average of 3.1-fold after both 5 and 10 weeks of NR supplementation, followed by 2.6-fold and 2.1-fold increases at 15 and 20 weeks, respectively. In the PBO-NR group, NAD⁺ levels remained near baseline during the initial 5 and 10 weeks of PBO. Following the switch to NR, levels increased by 2.6-fold and 2.1-fold after 5 and 10 weeks of supplementation, respectively. There were no between-group (NR vs PBO) differences in PSQI change scores or within-subject differences in the PBO-NR group. In the post-hoc analysis, when all participants were grouped, there was a significant association between PSQI change scores and NAD⁺ levels after 10 weeks of NR intake.

Conclusions: Increasing NAD⁺ levels may help improve sleep quality in people with long-COVID. Larger-scale clinical trials are needed to better understand the relationship between NAD⁺ levels and sleep quality.

Keywords: COVID-19, Sleep disturbances, nicotinamide riboside, long-COVID, NAD⁺

Disclosure: Nothing to disclose.

P162. CAPTURING THE PATIENTS' PERSPECTIVE: PATIENT-REPORTED OUTCOMES FROM IMPACT-1, A RANDOMIZED CONTROLLED PHASE 2 CLINICAL TRIAL OF TSND-201 (METHYLONE) FOR PTSD

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Background: Post-traumatic stress disorder (PTSD) is a debilitating psychiatric disorder affecting 13M in the US. The only FDA-approved pharmacotherapies are two selective-serotonin reuptake inhibitors (SSRIs, i.e., sertraline and paroxetine), which are slow-acting and remain ineffective for many. TSND-201 (methylone) is currently in clinical development for the treatment of PTSD, where it has shown rapid, robust and durable efficacy as well as a favorable safety profile in Phase 2 studies. In July 2025, TSND-201 was granted Breakthrough Therapy Designation from the FDA, highlighting the potential for TSND-201's superiority over currently available treatment options as well as the urgent unmet medical need for new PTSD treatments.

TSND-201 is a highly selective rapid-acting neuroplastogen that promotes neuroplasticity by increasing the release of serotonin, norepinephrine, and dopamine, without direct agonist/antagonist activity at serotonin 2A (5HT_{2A}) receptors. TSND-201 is non-hallucinogenic and has shown rapid, robust, and long-lasting

benefit in preclinical studies of PTSD-, depression-, and anxiety-like behaviors.

Methods: IMPACT-1 (ClinicalTrials.gov Identifier: NCT05741710) was a randomized, placebo-controlled Phase 2 study of TSND-201 in individuals with PTSD. Eligible participants were adults aged 18–65 who met the DSM-5 criteria for PTSD with at least 6 months of symptoms assessed by the Clinician-Administered PTSD Scales for DSM-5 (CAPS-5). Eligible participants had severe PTSD (CAPS-5 ≥ 35) at screening. Participants were randomized, received TSND-201 or placebo once per week for four-weeks without psychotherapy, and were followed for an additional 6-weeks after the last dose until Day 64 (end of study). Efficacy data were collected 2 days after each dosing session (Days 3, 10, 17, and 24) and during the follow-up period (Days 29, 36, 43, and 64). The mean (SD) baseline CAPS-5 total score was 45.8 (7.11) points in the TSND-201 group and 46.0 (5.42) points in the placebo group.

Results: We previously reported that the primary endpoint was met (i.e., the change in CAPS-5 total scores from baseline to Day 64). Specifically, TSND-201 demonstrated statistically significant greater improvement from baseline to Day 64 in CAPS-5 total score (LS mean [SE] change, -23.3 [2.84]) compared with placebo (change, -13.6 [2.95]), with a LS mean difference of -9.6 ($p = 0.011$). The most commonly reported TEAEs occurring in at least 20% of participants administered TSND-201 were headache, decreased appetite, nausea, dizziness, blood pressure increased, dry mouth, and insomnia.

Here, we present an analysis of patient-reported outcomes—the PTSD Checklist for DSM-5 (PCL-5), the Patient Global Impression of Severity (PGI-S), and the Patient Global Impression of Change (PGI-C)—which provide direct insights into patients' perceptions of their PTSD symptoms, and overall treatment progress, aspects that may not be fully captured by clinician-administered assessments alone. On Day 64, there was a statistically significant greater improvement in all three scores with TSND-201 vs. placebo. First, on the PCL-5, TSND-201 demonstrated a -28.5 (3.67) point change from baseline compared with -19.5 (3.81) in the placebo treated group, and a significant LS mean difference of -8.99 points (90% CI: -17.81 , -0.17 ; $p = 0.047$). Second, on the PGI-S, TSND-201 treatment resulted in a -1.0 (0.20) point change from baseline compared with placebo, which had a -0.5 (0.21) point change, resulting in a significant LS mean difference of -0.49 points (90% CI: -0.97 , -0.01 ; $p = 0.047$). Finally, PGI-C scores significantly improved with TSND-201 treatment, with 50.0% of participants endorsing the "Very much improved" or "Much improved" items compared with 15.4% of participants in the placebo group, resulting in a treatment difference of 34.6% (90% CI: 15.2%, 54.0%; $p = 0.004$).

Conclusions: Taken together, these results show that TSND-201 produces meaningful improvements across both clinician-administered and patient-reported outcome measures for PTSD, highlighting its potential as a robust and clinically impactful treatment option. These findings support the therapeutic potential of TSND-201 for PTSD and the importance of evaluating its efficacy and safety in a larger clinical sample in a Phase 3 trial.

Keywords: PTSD, methylone, CNS Clinical Trials, Patient Reported Outcomes, rapid-acting treatment

Disclosure: Transcend Therapeutics, Employee, Self.

P163. RAPID ANTIDEPRESSANT EFFECT OF INTRANASAL BPL-003 (5-METHOXY-N,N-DIMETHYLTRYPTAMINE) IN TREATMENT-RESISTANT DEPRESSION: A PHASE 2B DOUBLE-BLIND STUDY

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Background: BPL-003 is a novel intranasal formulation of 5-methoxy-N,N-dimethyltryptamine (5-MeO-DMT; mebufotenin benzoate), a short-acting psychedelic. Data from a Phase 2a open-label study in treatment-resistant depression (TRD; NCT05660642) suggested a single 10 mg dose of BPL-003 had acceptable safety and tolerability, with a rapid antidepressant efficacy that was sustained for up to 3 months [Roberts, C., et al., ECNP 2024: P2440]. The current Phase 2b study was conducted to assess the efficacy and safety of intranasal BPL-003 in patients with TRD, in an international, multicenter, double-blind, randomized, controlled, parallel design study, BPL-003-201 (NCT05870540).

Methods: TRD participants were aged 19 to 73 years, had failed between 2 and 5 adequate antidepressant trials in their current episode (MGH ATRQ criteria) and had moderate to severe depression. Participants who were not currently taking antidepressants, were randomized to receive a single dose of either 0.3 mg ("pseudo-placebo"), 8 mg or 12 mg BPL-003, administered intranasally, with 8 weeks of follow-up. Standardized psychological support was provided in the 2 weeks before, during and the 2 weeks after dosing.

The primary endpoint was change from baseline in the Montgomery-Åsberg Depression Rating Scale (MADRS) Total score for 12 mg compared to 0.3 mg BPL-003. Key secondary endpoints were change from baseline MADRS Total score at Day 8 (12 mg and 8 mg vs 0.3 mg) and Day 29 (8 mg vs 0.3 mg). MADRS assessments were conducted by blinded, independent raters. Other depression assessments included the Clinician Global Impression of Severity (CGI-S) and patient reported outcomes (Quick Inventory of Depressive Symptomatology [QIDS-SR-16]; Quality Life in Depression Scale [QLDS]). Safety was assessed with adverse events, safety blood tests, vital signs, ECG, and a readiness for discharge questionnaire.

Results: 193 participants were randomized (3:2:3), to receive a single dose of 0.3 mg ($n = 74$), 8 mg ($n = 46$) or 12 mg ($n = 73$) BPL-003 from 38 centers in 6 countries (Australia, Germany, Poland, Spain, UK, USA). There was a low discontinuation rate of 9.8%, with demographics and baseline characteristics similar between groups. Participants had a mean age of 41 years, 59% were female, 89% were white, and 38% were from the USA.

The overall Baseline MADRS score was 31.8, with 68% of participants classified as moderate depression (MADRS score < 35) at Baseline. Participants had failed a mean of 2.3 antidepressants in their current depressive episode, and 65% washed out of antidepressants during Screening.

The primary endpoint, change from baseline MADRS Total score at Day 29, 12 mg compared to 0.3 mg, was statistically significant (LS means difference [SE]: -5.3 [1.79]; $p = 0.0038$). There was a significant reduction in MADRS Total score from the first assessment at Day 2 until the last assessment at Day 57, for both 12 mg and 8 mg BPL-003 compared to the 0.3 mg dose. Clinically meaningful and statistically significant improvements in depression symptoms were also observed in the CGI-S and patient reported outcomes (QIDS-SR-16, QLDS) for the 12 mg and 8 mg doses, reinforcing the clinical relevance of these improvements.

BPL-003 was well tolerated at all doses. Treatment-emergent adverse events (TEAEs) were reported in 73%, 76% and 85% of participants, and drug-related TEAEs in 34%, 70% and 82% of participants, in the 0.3 mg, 8 mg and 12 mg arms respectively. The most common TEAEs were nausea, headache, administration site pain/discomfort, anxiety, blood pressure increased, vomiting and insomnia. There were two serious adverse events, both of which

were determined to be not related to BPL-003; moderate depression in the 0.3 mg arm and moderate agitation in the 8 mg arm. The majority (>99%) of TEAEs were mild or moderate and reported on dosing day. Transient increases were observed in blood pressure and heart rate; there were no clinically significant safety concerns observed in ECG or safety blood tests.

Participants were deemed to be ready for discharge at a median time of <2 h following all dose levels.

Conclusions: This is the largest, controlled, international study to date of 5-MeO-DMT. It provides further evidence for the potential benefit of intranasal BPL-003 in the treatment of TRD. A single dose of BPL-003, in patients with moderate to severe symptoms, demonstrated clinically relevant improvements in MADRS score, with rapid onset of effect that was sustained for at least 8 weeks. This data supports further investigation of BPL-003's antidepressant properties in a Phase 3 program.

Keywords: psychedelic drugs, 5-MeO-DMT, treatment-resistant depression, Major Depressive Disorder (MDD)

Disclosure: Beckley Psytech, Contracted Research, Self.

P164. NEUROFLEX IMPROVES EVENT-RELATED POWER IN PATIENTS WITH CANCER-RELATED COGNITIVE IMPAIRMENT

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Background: Advances in cancer treatment are resulting in a growing number of cancer survivors. A distressing and increasingly recognized long-term sequela reported by cancer patients and survivors is cancer/chemotherapy-related cognitive impairment (CRCI). CRCI most often involves difficulties with executive functions and other cognitive processes following cancer treatment. Although CRCI has been reported by patients for decades and is well-supported by empirical evidence, survivors frequently describe a lack of recognition or validation of their cognitive symptoms within clinical settings. As a result, many survivors are left to navigate the long-term cognitive effects of cancer treatment without adequate support. There remains a critical need for accessible, evidence-based interventions tailored to this population. A scalable, low-cost, and well-tolerated digital intervention offers a promising and innovative solution for addressing persistent CRCI. We previously reported the feasibility and acceptability of NeuroFlex, a digital cognitive enhancement tool, in cancer survivors with CRCI. NeuroFlex is theoretically founded in animal models of induction of neuroplasticity in the aging brain and is designed to target the cognitive functions commonly affected in CRCI, specifically executive dysfunction, through the induction of neuroplasticity. The present study examined whether 6-weeks of NeuroFlex training in participants with persistent CRCI influenced functional brain activity, as measured by EEG, during a go/nogo task and an incidental memory task. We hypothesized that NeuroFlex would improve event-related neural activity across both tasks.

Methods: This study followed a single-arm, open-label design with cognitive, behavioral, incidental memory, and go/nogo EEG tasks completed pre- and post-intervention. Participants were screened to exclude individuals with evidence of clinically manifest cognitive impairment or depression. Study participants had undergone treatment with systemic chemotherapy within the last 1–8 years and endorsed persistent CRCI subjective complaints (as measured by the Cognitive Complaint Index). Twenty-one female (N = 21) breast (n = 17), ovarian (n = 3), and endometrial

cancer (n = 1) survivors completed 6-weeks of NeuroFlex training. The goal for training was 8hrs/week and 45 hours total over 6 weeks. EEG activity was recorded using a 256-channels and event-related power was extracted in 4 frequency bands between 2 and 30 Hz. A go/nogo task was used to evaluate changes in response inhibition or inhibitory control, which are executive functions. A passive incidental visual memory paradigm was used to evaluate changes in implicit recognition memory.

Results: Participants completed an average of 7.42 h of nCCR training per week, an average of 44.5 (± 1.01) hours total, and had 100% completion (n = 21) for participants (mean age = 56.45 ± 11.03 years) who received the intervention. For the go/nogo task, the analyses of evoked power over the frontal ROI revealed a significant session-by-trial interaction observed between 400–600 ms in the alpha frequency band ($F(1, 20) = 5.74$, $p < 0.03$, $\eta^2_p > 0.22$). This interaction was driven by a reduction in alpha desynchronization for go trials compared to nogo trials during the post-treatment session. This change indicates that post-treatment, participants processed frequent go trials with less effort, while maintaining the neural resources necessary to engage in more effortful inhibitory processing for the less frequent nogo trials. For incidental memory task, over the parietal ROI, there was a significant session-by-trial interaction for the alpha power from 0 to 300 ms (all $p < 0.04$, $\eta^2_p > 0.19$). This interaction was driven by the reduction in alpha desynchronization for novel vs repeat images in the post-treatment session. This pattern suggests greater inactivation for non-salient stimuli (novel images), while repeated images engaged in more salient, task-relevant processing. In addition, alpha power changes correlated with improved self-reported mood and performance on cognitive tasks.

Conclusions: The results of the present study indicates that 6-weeks of NeuroFlex training resulted in functional shifts in cortical activation patterns in cancer survivors with persistent CRCI. The intervention was exceptionally well tolerated, with 100% of participants who started the program completing it. NeuroFlex not only improved behavioral performance across memory and executive function tasks but also optimized neural processing, as evidenced by task-specific changes in alpha power. These results align with theoretical frameworks suggesting that targeted cognitive training can induce neuroplasticity in distributed brain networks. These results highlight the utility of EEG as a tool for capturing subtle changes in neural dynamics that may not be immediately apparent through behavioral measures alone. Furthermore, the observed correlations between neural changes and improvements in mood and cognition underscore the interconnected nature of cognitive and emotional processes, suggesting NeuroFlex may have benefits that extend into multiple aspects of daily functioning. As CRCI remains a challenging and under-addressed issue, the present study emphasizes the importance of developing accessible, scalable interventions capable of producing measurable neural and behavioral benefits in cancer survivors with CRCI.

Keywords: Digital Medicine, cognitive remediation, EEG, breast cancer, Chemobrain

Disclosure: Nothing to disclose.

P165. EFFECTS OF TRANSCRANIAL DIRECT CURRENT STIMULATION (TDCS) ON SYMPTOMS AND BRAIN CHANGES IN PATIENTS WITH AUTISM

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Background: Patients with autism spectrum disorder (ASD) have severe social deficits and behavioral and educational treatments have limited effectiveness. There is preliminary evidence that tDCS can improve some symptoms in patients with ASD. Previous studies have shown that synaptic E/I ratios are significantly elevated in the medial prefrontal cortex (mPFC) of autism-like mouse models, and that such excessive elevation results in severe social impairment phenotypes and that abnormalities in mPFC may be associated with core symptoms of ASD. This background provided a rationale for a study of the effects of tDCS targeting the mPFC in children with ASD.

Methods: This was a randomized double blind study of the effects of tDCS in patients with a diagnosis of ASD. 36 patients with ASD in Changsha, China were randomized to receive either 1.5 mA anodal tDCS or Sham tDCS for 20 min 2 times/day for 7 days delivered to the mPFC. Resting state EEG recordings were analyzed to assess changes in power spectral density (PSD) slope and the functional excitation-inhibition balance (fE/I) ratio. The primary clinical outcome measure was change in the Ohio State University Autism Rating Scale for DSM-5 (OARS-5) and there were several secondary clinical and safety measure scales. Analysis of the clinical data used linear mixed models in Stata 19 and SAS 9.4. Correlations were used to assess relationship of clinical change and EEG measure changes.

Results: The active tDCS group showed significant greater decrease, compared to sham, in OARS total score ($P < 0.01$) and social functioning sub-score ($P < 0.03$) after the 7 days to tDCS treatment and some of the differences in OARS scores persisted at an assessment 3 weeks later. There was a significant increase in slope ($p = 0.004$) and a significant decrease in fE/I ($p = 0.011$) from baseline to post week1 in the active tDCS group, with no significant changes observed in the sham group. Correlational analyses indicated that reductions in fE/I ($r = 0.377$, $p = 0.044$) and increases in slope ($r = -0.447$, $p = 0.015$) were significantly associated with improvements in OARS-5 social functioning scores.

Conclusions: Our results suggest that tDCS to mPFC may be effective in reducing some symptoms associated with ASD, and induce changes in excitation-inhibition balance in the brain as analyzed from resting state EEG analyses. Correlations suggest that the changes in OARS scores may be influenced by changes in fE/I and slope brain measures.

Keywords: autism Spectrum Disorders, TDCS, Social Deficits, Medial Prefrontal Cortex

Disclosure: Nothing to disclose.

P166. A SMARTPHONE-BASED INTERVAL TIMING TASK FOR ASSESSING COGNITIVE FUNCTION

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Background: Cognitive deficits are common in a range of neuropsychiatric disorders including schizophrenia, bipolar disorder, autism, and ADHD. These deficits include impairments in working memory, attention, planning and temporal processing. There are currently no treatments for cognitive dysfunction. We use interval timing, to study cognitive function in humans and in rodents. Interval timing refers to the ability to perceive, estimate, and produce time intervals which we study in the range of seconds to minutes. Interval timing is a fundamental cognitive process which many species use to track and make temporal predictions about events in their environment.

Our group and others have identified a circuit in the brain that supports multisecond Interval Timing that includes cortico-striatal

systems and the cerebellum. We explore this circuitry in humans and in rodents with a modified operant task and have reported interval timing impairments in patients with schizophrenia and bipolar disorder. Interval timing in humans could provide a novel tool for assessing cognitive dysfunction.

Identifying the precise neural circuitry involved in interval timing presents a significant challenge due to the variability in human experience. Intersubject heterogeneity has complicated our efforts to map consistent neural mechanisms necessary for temporal cognition. To overcome these challenges, we designed an interval timing task to be administered via smartphone or personal electronic device. Our goal with using a digital tool is to increase participation accessibility and define demographic and environmental factors that may account for variability in timing performance. These data will guide our understanding of temporal cognitive processing across neuropsychiatric conditions.

Methods: For this study, we developed the interval timing task for application on the BlinkLab smartphone application developed at Princeton University. We are currently validating and optimizing the application for use by collecting data from all interested participants at the University of Iowa and surrounding community. We are not excluding participants based on age, sex, diagnosis, or medication status. On the application, participants receive instructions to begin a time estimation task. Participants are asked to avoid tracking time by counting in head or foot tapping, and when they see a 3 or a 9, they are to close their eyes for that duration of time and open them afterwards. They are allowed to blink normally throughout the task. We measure time estimation based on accuracy (i.e., absolute deviation from target interval) and variability (i.e., standard deviation across trials). Participants also complete brief demographic and behavioral questionnaires (e.g., age, sleep, education, diagnosis, medication status, sleep, and exercise).

Primary analyses will use linear mixed-effects models to examine trial-by-trial accuracy and variability with each participant as a random effect, and covariates (e.g., age, sex, sleep, diagnosis) as fixed effects. Test-retest reliability for identically timed trials will be assessed with intraclass correlation coefficients (ICC) and Bland-Altman plots. Associations with covariates will use Pearson or Spearman correlations depending on distributional assumptions. Model assumptions will be verified and reported with effect sizes, confidence intervals, and p-values ($\alpha = 0.05$).

Results: Our goal is to identify demographic and behavioral factors that contribute to heterogeneity in interval timing accuracy and variability. Preliminary analyses indicate that the BlinkLab interval timing task reliably captures multisecond interval timing performance, with trial-by-trial estimates showing consistent within-participant patterns. Based on prior literature, older age is expected to be associated with decreased timing accuracy, shorter sleep with increased variability, and lower physical activity with reduced accuracy and greater variability. These predictions will be formally tested in our study to evaluate the generalizability of prior findings and the translational utility of the BlinkLab task.

Conclusions: Building on prior interval timing findings from our lab, this task provides a novel timing as a window into cognitive function. Our findings demonstrate the feasibility of a smartphone-based interval timing task in humans and offer an accessible tool to quantify interval timing as a cognitive phenotype. By translating circuit-level insights from animal models to human clinical studies, with increased accessibility, this translational paradigm supports future research into cognitive markers of neuropsychiatric disorders.

Keywords: interval timing, cerebellum, digital assessment, Cognition, Translational Neuroscience

Disclosure: Nothing to disclose.

P167. A NOVEL CHARACTERIZATION OF PROSPECTIVE SELF-CONTROL COST ESTIMATES

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Background: Failures to exert control over eating behavior remains a significant challenge for individuals. One proposed reason for these failures is because individuals cannot meet the cognitive demands ('costs') required to adequately deploy control. Here, across 3 studies, we characterize how healthy dieters estimate the cognitive cost of exerting control over varying levels of food temptation and probe the motivational, neural and computational mechanism underlying these costs.

Methods: Healthy dieters [Study 1: $n = 96$ (behavioral); Study 2: $n = 93$ (behavioral); Study 3: $n = 50$ (fMRI)] completed a self-control decision task during which they reported trial-by-trial willingness-to-pay (WTP) to avoid foods that varied on temptation level, quantity and food exposure time. One trial was randomly selected at the end and played out for real. Study 2 included a within-subject manipulation that tested cost changes when foods were available vs. unavailable to eat. Study 3 was conducted in the fMRI scanner and analyzed using a parametric modulator of WTP value. Across studies, choice data was fit to a computational model that described individuals' cost function for self-control.

Results: Linear mixed-effects models revealed that temptation level, quantity and exposure time were all positively associated with higher WTP to avoid control (all p 's < 0.001). Further, WTP was significantly higher when foods were available (vs. unavailable) for consumption ($p < 0.0001$). Computational modeling revealed that the algorithmic process underlying the increase in costs scaled in a multiplicative manner. Finally, higher WTP yielded increased activation in frontopolar cortex and dorsolateral PFC pointing to a central role in these brain regions in prospectively estimating the perceived cost of self-control.

Conclusions: Our findings indicate that the cognitive cost of self-control scales with attributes that impose higher self-control demands, and that increased temptation amplifies these costs disproportionately over time. Our fMRI results point to a neural circuit consistent with prospective decision-making and control cost estimation. Acquiring a better understanding the neurocomputational basis of these cost estimates may provide potential neural targets to help improve the success of prospective control strategies.

Keywords: self-control, decision-making, Cognitive effort

Disclosure: Nothing to disclose.

P168. NEURAL MECHANISMS OF CONTROL OVER AUDITORY VERBAL HALLUCINATIONS

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Background: Hallucinations are a core feature of psychosis and can be disabling and distressing. However, many people with hallucinations function well in spite of—and even because of—these experiences. The degree to which their onset and offset is under voluntary control has emerged as a promising predictor of

functioning among voice-hearers, but the neural mechanisms underlying this ability remain unclear. Many voice-hearers report an ability to regulate or suppress voices, implicating prefrontal inhibitory networks. Identifying the neurobiological substrates supporting this control is vital for understanding symptom heterogeneity and developing targeted interventions to enhance control and functioning in voice-hearers.

Methods: Control over voice-hearing was assessed using the Yale Control Over Perceptual Experiences (COPE) scale, a validated instrument that reliably quantifies the degree and methods of voluntary control. Sixty participants with AVH (30 high-control, 30 low-control as classified by COPE scale scores) completed an fMRI paradigm integrating symptom-capture and volitional control. The task included three runs of seven 40-s blocks instructing participants to suppress voices ("turn off"), engage voices ("turn on"), or remain neutral ("do nothing"), with voice onset and duration indicated by button press and hold. A final control block required continuous button pressing for the average reported voice-hearing duration without modulation instructions, allowing separation of perceptual neural signals from motor reporting activity. Images were acquired using a 3T Siemens Prisma scanner (TR = 800 ms or 1s, TE = 37ms, voxel = $2 \times 2 \times 2$ mm), preprocessed via fMRIPrep, and harmonization of scanning parameters used the COMBAT algorithm. Statistical maps were thresholded using a false positive rate control of $\alpha = 0.05$, with a minimum cluster size threshold of 50 mm³.

Results: Participants with greater control over voice-hearing demonstrated improved inhibitory performance, reflected by reduced AVH button press durations during suppression blocks (mean difference: $\Delta\mu = -2.38$ s, 95% CI [-3.00, -1.77], $p = < 0.001$). They also showed lower button press durations during neutral blocks (mean difference: $\Delta\mu = -1.12$, 95% CI [-1.86, -0.38], $p = 0.005$). At the neural level, when comparing inhibition versus control button press conditions, high-control voice-hearers showed greater engagement of frontal executive networks, particularly in the superior frontal gyrus (MNI: 20, -18, 46; 435 mm³, $Z = 3.62$) and inferior frontal gyrus (46, 15, 6; 288 mm³, $Z = 3.74$), regions critical for executive function and inhibitory control.

High versus low control voice-hearers showed distinct patterns of activation during voice hearing periods, with differences centered on the medial orbitofrontal cortex (-1, 30, -22; 1790 mm³, $Z = 4.05$) and extending into perceptual processing regions in temporal and fusiform areas (Z -scores 3.29–4.26). Voice elicitation compared to button press controls engaged subcortical and parietal regions, particularly the thalamus and superior parietal areas (Z -scores 3.76–3.97), suggesting differential recruitment of attention and sensory integration networks.

Conclusions: Individuals with greater control over their voices demonstrated both superior behavioral inhibition and characteristic neural activation patterns. During active suppression, these participants showed increased engagement of frontal executive regions (superior and inferior frontal gyri), along with heightened activation in the medial orbitofrontal cortex and perceptual processing areas during voice-hearing periods. These findings have important clinical implications: individuals with greater ability to control their voices often experience less distress and impairment as well as better outcomes. Understanding the neural basis of hallucination control can also guide personalized treatment strategies and identification of individuals at risk for more persistent or disabling symptoms.

Keywords: Auditory hallucinations, cognitive control network, Functional MRI (fMRI)

Disclosure: Nothing to disclose.

P169. DECISION-MAKING AND REWARD IMPAIRMENTS IN ADHD ADOLESCENTS

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Background: The role of reward system dysfunction as a possible etiological factor in Attention-Deficit/Hyperactivity Disorder (ADHD) has increasingly become the focus of research. This study examined differences in learning style in ADHD-diagnosed adolescents using the Multi-Stage Decision-Making task (MSDM) that can discriminate between “model-based” and “model free” response styles. Our aim was to compare ADHD versus non-ADHD response styles by examining response profiles as well as with an innovative application of Drift Diffusion Modeling (DDM) to parameterize how both groups aggregated information to make their first-stage choices. Compared to non-ADHD, we predicted ADHD-diagnosed adolescents would show greater evidence of being model-free learners due to theorized ADHD reward sensitivity. A secondary goal was to learn whether any response-style abnormalities could be explained by either rapid, superficial information aggregation, or an inefficient, slower, and more deliberative decision-making process.

Methods: 149 ADHD and 130 non-ADHD ($N = 279$) adolescents completed the MSDM (or “two-stage” learning task). In this task, participants make a first-stage choice that only sometimes leads to one of two second-stage screens (70% common/30 rare transitions). At this second stage, participants again make a forced choice between two stimuli and learn if they will receive a reward. Optimizing outcome can be achieved using a model-based approach that tracks slowly-shifting reward probabilities to second-stage stimuli over 200 trials so one can make first-stage choices that most often reach the set of stimuli with the highest reward likelihood. In contrast, a model-free style relies more on immediately prior reward delivery. Response styles were examined by quantifying the probability of first-stage “stays” (when participants repeated the same choice) as a function of previous trial outcome, separately for common versus rare transitions. The Hierarchical Sequential Sampling Modeling (HSSM) Python toolbox was used to quantify DDM ‘ v ’ (drift rate), ‘ a ’ (boundary separation), and ‘ z ’ (response bias) parameters. Mixed-effects Bayesian linear models tested these parameters for significant main and interaction effects of diagnosis, prior trial outcome, and prior trial transition type.

Results: Both ADHD and non-ADHD groups’ response style had hallmarks of using a combination of model-based and model-free strategies. However, ADHD had lower first-stage stay proportions following rewarded common transition trials ($p = 0.02$) and non-rewarded rare transition trials ($p = 0.08$). Analysis of DDM parameters found ADHD had a slower ‘ v ’ drift rate (Bayesian $P = 1.0$) and higher ‘ a ’ decision threshold ($P = 0.99$) compared to non-ADHD. ADHD also had a decision bias not seen in non-ADHD ($P = 0.98$). These diagnostic group differences were unaffected by whether the previous trial transition was common or rare. However, prior trial reward delivery did influence how information was aggregated on the next trial. While reward generally increased drift rate on subsequent trials, this effect was less in ADHD ($P = 0.99$). Similarly, the increase in ‘ a ’ boundary separation seen in both groups after rewarded trials also was significantly less prominent in ADHD ($P = 0.99$).

Conclusions: Contrary to predictions, this study did not find strong evidence that ADHD-diagnosed adolescents relied more greatly on model-free learning styles. However, the lower ADHD stay proportions reflected a response profile that was observed in prior studies to be less developmentally mature. This study’s DDM

analyses offer new insights into the nature of information processing that underlie these subtle group differences. The DDM results revealed that ADHD participants needed more information (‘ a ’), that was aggregated at a slower rate (‘ v ’), and from a slightly biased starting point (‘ z ’). The DDM findings can be interpreted as generally less effective learning and decision systems in ADHD while performing this complex task. Such an interpretation might be related to neurobiological factors in ADHD that lead to inefficiencies when discriminating neural signals from aggregate decision-making system noise. Also, ADHD-diagnosed adolescents’ decisions were less influenced by reward information from immediately prior trials. That interaction effect aligns with the group findings, as decreased reward sensitivity also can result from greater decisional noise. This suggests that reward outcomes are not as effective in ADHD when mobilizing cognitive resources during the learning process. To our knowledge, this is the first study comparing ADHD to non-ADHD adolescents using the MSDM task, as well as the first application of DDM to an MSDM task to give insights into cognitive mechanisms that might underlie each group’s response style. The presentation will further elucidate the impact of MSDM task factors on the DDM parameters between groups. Additional testing of these data (e.g., to explore second-stage decisions) is currently underway to elaborate upon this foundation of novel findings.

Keywords: ADHD, Drift-Diffusion Modelling, Reward, Adolescents

Disclosure: Nothing to disclose.

P170. GENETIC CONTRIBUTIONS TO WORKING MEMORY ACTIVATION IN EARLY ADOLESCENCE - LINKS TO PSYCHOPATHOLOGY AND COGNITION

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Background: Engagement of working memory neural circuitry has been hypothesized to partially mediate the effects of genetic risk for psychopathology on mental health outcomes. We probe this hypothesis using a recently developed ‘neural signature’ of working memory, which reflects individual differences in whole-brain engagement of task-relevant regions during performance of a working memory fMRI task.

Methods: Analyses included $N = 4737$ participants ($n = 7253$ observations, ages 9–13, 53% male) from the first two imaging waves of the ABCD Study® (5.1 release) whose genomic ancestry resembled those of European reference samples. Polygenic scores (PGS) for 15 mental health-related traits (e.g., cognition, schizophrenia, ADHD, bipolar disorder, risk-taking, addiction risk, anxiety, depression, etc.) were generated using PRS-CS. The neural signature was derived by training a whole-brain elastic net decoder to distinguish between the high and low working memory loads of the Emotional N-back fMRI task in split-half analyses. Prior work has shown that the neural signature is more reliable and a more valid indicator of working memory ability than activation in individual regions, and it has been shown to generalize across scanners and samples. Analyses examined an individual difference score calculated as the difference in decoder output between each participant’s 2-back and 0-back activation maps, reflecting within-subject changes in engagement of task-relevant regions. Mixed-effect models adjusted for site, family, and individual ID as random-effects, and fixed effects (e.g., age, age², sex, 10 ancestry principal components, in-scanner head motion).

Results: Significant ($p < 0.05$ fdr) PGS associations with the working memory neural signature were found with cognition

($\beta = 0.11$, $p = 3 \times 10^{-19}$), schizophrenia ($\beta = -0.05$, $p = 9 \times 10^{-5}$), and ADHD ($\beta = -0.06$, $p = 9 \times 10^{-6}$). Participants with greater cognition PGS had more activation of task-relevant regions during working memory; participants with greater schizophrenia and ADHD PGS show reduced activation of task-relevant regions. Within-between family analyses ($N = 1389$ related participants, $n = 2156$ observations) supported direct genetic effects for cognition (within-family $\beta = 0.11$, $p = 0.008$; between-family $\beta = 0.14$, $p = 1.7 \times 10^{-6}$), while confounding (e.g., passive gene-environment correlations) could not be ruled-out for others, likely due to the smaller size of the family sub-sample. As in prior reports, related measures of psychopathology (attention problems and severe psychotic-like experiences) and total cognition were associated in a directionally consistent manner with the neural signature ($|\beta| \geq 0.07$, $p < 0.05$ fdr) and the corresponding PGS ($|\beta| \geq 0.11$, $p < 0.05$ fdr). As such, the neural signature significantly partially mediated the association between PGS and cognition, attention problems, and severe psychotic-like experiences ($p < 0.05$ fdr). Post-hoc analyses examining sex found no evidence for moderation.

Conclusions: Neural activation during working memory is a plausible partial mediator of the association of genetic risk with cognitive and mental health outcomes. Within-between family analysis results are consistent with a model in which cognition genetics causally influence working memory neural function (i.e., within-family effects were significant). A limitation of this work is that analyses are restricted to participants whose genomic ancestry is similar to that of the samples in the genome-wide association studies that were used to generate PGS. Differential neurodevelopment of the circuitry supporting working memory may be a contributing mechanism through which genetic liability to psychopathology and cognition manifests in adolescence. Future work will test for longitudinal mediation and associations with longitudinal growth trajectories.

Keywords: working memory fMRI, Imaging-genetics, ABCD study, machine learning, ADHD

Disclosure: Nothing to disclose.

P171. A MULTIVARIATE DATA FUSION OF NT2* BRAIN IRON CONCENTRATION AND DIVERSE CLINICAL AND COGNITIVE PROFILES IN ATTENTION-DEFICIT/HYPERACTIVITY DISORDER

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Background: Normalized T2*-weighted imaging (nT2*) is a noninvasive index of brain tissue iron concentration believed to quantify dopamine system function in brain regions with high D2 receptor or dopamine transporter densities. Recent studies have begun to replicate that persons diagnosed with Attention-Deficit/Hyperactivity Disorder (ADHD) have abnormal nT2*-measured values, particularly in subcortical regions that typically have the highest concentration of brain iron. However, one important consideration that hampers advances in this line of research is a general lack of guidance as to whether nT2* values are clinically significant. In this study, we build upon our prior findings of ADHD adolescent nT2* abnormalities by characterizing the complex multivariate associations between whole-brain nT2* and a diverse battery of clinical, cognitive, reward, and psychosocial function measures, then testing to see how those relationships differ between ADHD and non-ADHD.

Methods: Time-averaged nT2* is calculated from BOLD T2* fMRI data because T2* relaxation rates are higher in the presence

of brain iron. By averaging across many T2* volumes small BOLD task-correlated signal changes that typically would be of interest in conventional fMRI analysis are discarded, so instead mean signal susceptibility effects due to iron can be retained with high signal-to-noise. 149 ADHD and 143 non-ADHD adolescents underwent echo planar imaging fMRI for approximately 45 min while they completed various fMRI tasks. Data were prepared for analysis using Human Connectome Project processing, including ICA-FIX denoising and MSMAll surface-based registration steps. Prior to timeseries averaging, any volume whose framewise displacement value was >0.50 was censored. Also, each BOLD image was normalized by dividing it by the mean its voxel values. For all participants, a large battery of dozens of clinical, cognitive, reward, and psychosocial functioning measures was collected. There were 177 variables for analysis after screening all variables to eliminate any redundant measures with problematic collinearity. Because values were missing for 6.49% of the data, nonlinear single imputation using a classification and regression tree approach was used to avoid listwise deletion of entire cases. Finally, the variables in this feature set were z-transformed to ensure none would have an outsized influence in the subsequent "data fusion" step due to any disproportionate scaling. Multi-set Canonical Correlation Analysis with joint Independent Component Analysis (MCCA+jICA) combined the nT2* feature with the Clinical/Cognitive feature. MCCA+jICA identifies multivariate relationships between the two features that are systematically conserved across participants. The two feature sets were fused into eight canonical variates (CV). Two-sample t tests then tested for differences between ADHD and non-ADHD on these CVs' mixing coefficients to identify if the complex relationships were expressed differently in each study group.

Results: Four CVs significantly differed between ADHD and non-ADHD – three for which the nT2* feature differed (all p 's < 0.002) and all four where the Clinical/Cognitive feature differed (all p 's < 0.005). Interestingly, a comparable set of over a dozen variables out of 177 had the highest loadings ($z > 1.65$) on all 4 CVs. These variables were a mix of temporal estimation, N-back working memory, Go/NoGo, CPT performance consistency, delay discounting, CBCL measures of school competence or internalizing problems, and peer trust. For one CV, stronger or more positive scores were linked to lower nT2* (more brain iron, presumably higher DA function) in caudate, nucleus accumbens, hippocampus, and right ventrolateral and frontopolar prefrontal cortex (PFC). This profile was expressed more strongly in non-ADHD ($p = 0.005$). Visualization of this CV's nT2* feature group differences revealed non-ADHD had higher brain iron in subcortical and brainstem areas. In all three other CVs, a negative profile of the Clinical/Cognitive variables predicted less brain iron in different profiles: (i) largely in brainstem and vmPFC, (ii) in many lower diffuse cortical regions, or (iii) prominently in ventrolateral and ventromedial PFC regions.

Conclusions: To our knowledge, this is the first demonstration that relative levels of nT2*-measured brain iron predicts cognitive and clinical individual differences. This study also extends prior findings of lower ADHD brain iron by linking abnormally low subcortical and brainstem brain iron in ADHD to a profile of diminished cognitive and clinical functioning. Study strengths include the large sample size, the high resolution of nT2* measurements, the diverse breadth of the Clinical/Cognitive test score battery, and the rigorous MCCA+jICA data fusion approach. The presentation will further detail specific group difference profiles and explore relationships with other ADHD-relevant factors (e.g., symptom severity, recent psychostimulant medication use, etc.).

Keywords: ADHD, brain iron, fMRI, nT2*, multivariate data fusion

Disclosure: Nothing to disclose.

P172. ANHEDONIA AND EARLY LIFE UNPREDICTABILITY DISTINCTLY SHAPE MOTIVATIONAL CONFLICT RESOLUTION IN ADULTS

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Background: The decision-making that occurs under approach-avoidance motivational conflict promotes survival in environments where animals must balance their need for rewards with the sometimes-competing need to avoid catastrophic threats. There is growing evidence from rodent and human experiments that brain circuits underlying motivational conflict resolution may be altered by childhood adversity, specifically, early life unpredictability (ELU), and may be differentially affected in anhedonia, a transdiagnostic construct characterized by the reduced ability to experience pleasure or interest in activities that are typically rewarding. However, conflict resolution consists of a number of component processes and it is currently unknown which specific component(s) are altered in association with ELU or anhedonia.

Methods: We developed a novel task, "Revenge of the Zorn," a gamified version of a motivational conflict challenge adapted from Korn and Bach (2019). Participants played as space explorers trying to maximize their chance of surviving as many planets as possible without running out of fuel (i.e. following optimal policy). On each planet they were asked to decide between 1) playing a cardgame with a trial-varying, explicit chance of gaining fuel, losing fuel, or being robbed by a Zorn alien, versus 2) paying one fuel point to skip the trial and wait for the next trial (avoidance strategy). The total sample consisted of 111 participants, including: a web-based cohort (Prolific; $n = 98$, 52 Female, 57 Male, 2 "Other"; 32.7 ± 11.2 years), and an fMRI cohort ($n = 13$, 4 Female, 8 Male, 2 "Other"; 28.9 ± 9.47 years). ELU was measured using a latent factor decomposition of the Questionnaire of Unpredictability in Childhood (QUIC), validated to correspond to performance differences in a previous study of reward-guided decision-making by our group. Anhedonia was measured by the Mood and Anxiety Symptoms Questionnaire - Anhedonic Depression Subscale (MASQ-AD).

Results: ELU and anhedonia had opposing effects on overall gameplay, despite a positive correlation between them ($r = 0.13$, $p = 0.1$). Participants with higher ELU survived fewer planets ($r = -0.25$, $p = 0.008$), consistent with their choices being less likely to follow the optimal policy ($r = -0.28$, $p = 0.003$). This was attributed to greater avoidance in response to threat ($r = -0.22$, $p = 0.02$), despite being more likely to forage overall ($r = 0.2$, $p = 0.035$). In contrast, participants with high anhedonia survived more planets ($r = 0.28$, $p = 0.003$) and were more likely to follow the optimal policy ($r = 0.28$, $p = 0.003$). Interestingly, a high/low sub-group analysis of ELU and anhedonia was conducted and revealed that individuals with low ELU and high anhedonia performed the best in the game, with the most planets survived (median = 63 planets) and greatest adherence to optimal strategy (Kruskal-Wallis rank sum test, $\chi^2(3) = 18.7$, $p < 0.001$, $\chi^2(3) = 16.5$, $p < 0.001$, respectively). On the basis of previous work suggesting that ELU modulates valenced learning, and observed differences in learning from negative, versus positive, outcomes in anhedonia, we examined how participants responded to surprising negative events. Negative feedback learning was operationalized as the effect of surprising, negative trials on later choice (where a robbery by the Zorn alien occurred but the probability of robbery had been low). Participants with higher levels of ELU were more likely to play their card following surprising robbery, where participants with elevated anhedonia were more likely to choose "wait" ($t = 3.0$, $p = 0.002$; $t = -2.81$, $p = 0.004$).

Conclusions: Early life unpredictability and anhedonia have surprising, opposing effects on motivational conflict resolution behaviors in our task: ELU seemed to impair motivational conflict resolution, and anhedonia seemed to enhance it. A high/low sub-group analysis provided further evidence of a buffering effect of ELU on enhancement by anhedonia in this decision-making process. Following a surprising robbery (threat) result, anhedonia increased participants' probability of choosing to wait on the next planet, where increasing ELU made it less likely participants would choose to wait. This observation supports the idea that individual differences in overall gameplay derive from distinct responses to negative feedback, even under complete information. Previous studies have found diminished approach and enhanced avoidance behavior in depression cohorts related to decreased reward sensitivity, but here, we investigate anhedonia in a non-clinical population. This sample may provide evidence for an adaptive feature of anhedonia at subclinical levels for survival decision-making under motivational conflict that is enhanced with predictable childhoods.

Keywords: approach-avoidance conflict, anhedonia, early life unpredictability

Disclosure: Nothing to disclose.

P173. FLUID COGNITION CHANGES FOLLOWING REPEATED KETAMINE INFUSIONS IN MOOD DISORDERS: PRELIMINARY RESULTS

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Background: Patients with mood disorders frequently experience cognitive deficits. These deficits are associated with markers of severity of disease, including inflammation. Some studies show that ketamine, a glutamatergic modulator with rapid-acting antidepressant effect, lowers inflammation and improves cognitive performance in individuals with mood disorders. This ongoing study aims to assess fluid cognition (FC) in subjects with bipolar disorder (BD) and major depressive disorder (MDD), the correlation between fluid cognition and C-reactive protein (CRP) as well as depression symptoms, and changes in FC after one or more ketamine infusions.

Methods: Twelve individuals with mood disorders (5 BD and 7 MDD; mean age = 36.91 ± 12.36 years; 7 female) completed the NIH Toolbox Fluid Composite (NIH-TBFC), the Montgomery-Åsberg Depression Rating Scale (MADRS), the Young Mania Rating Scale (YMRS), and a baseline blood draw for CRP. All participants had at least mild depression with baseline MADRS scores > 12 [mean = 22.58, standard deviation (SD) = 5.77]. Participants did not have (hypo)manic symptoms, with YMRS < 8 (mean = 3.16, SD = 2.69). Seven participants received repeated subanesthetic intravenous ketamine infusions (up to five infusions for 3 weeks; 0.5 mg/kg over 40 min) and repeated the NIH-TBFC and MADRS after the first and last infusions. The remaining participants comprised the control "no-intervention" group and repeated the assessments to control for practice effects. The NIH-TBFC evaluates processing speed [Pattern Comparison Test (PCT)], working memory [List Sorting Working Memory (LSWM)], executive function [Dimensional Change Card Sort (DCCS)], attention [Flanker Inhibitory Control and Attention (FICA)], and episodic memory [Picture Sequence Memory (PSM)]. Linear mixed models examined the effect of condition (baseline, no-intervention, post-infusion 1, post-infusion 5) on age-adjusted fluid composite scores and individual cognitive domains, controlling for diagnosis, MADRS, and within-subject variability.

Pearson correlations assessed associations between baseline MADRS and baseline CRP with cognitive domains.

Results: Ketamine was associated with significant improvement in FC [$F(3, 13.25) = 8.59, p = 0.002$]. Specifically, differences were observed between baseline and post-infusion 1 ($p = 0.027$) and between baseline and post-infusion 5 ($p = 0.002$), with no difference between baseline and follow-up in the no-intervention group ($p = 0.20$). MADRS scores did not change significantly after ketamine [$F(3, 16.2) = 1.03, p = 0.40$]. Regarding individual cognitive domains, PCT improved with ketamine [$F(3, 13.34) = 19.83, p < 0.001$], although there was no difference between no-intervention and post-infusion 1 ($p = 0.65$) or post-infusion 5 ($p = 0.70$). There was a trend toward improvement in LSWM with ketamine [$F(3, 13.4) = 2.33, p = 0.1$], particularly between baseline and post-infusion 5 ($p = 0.09$). Ketamine did not significantly affect DCCS [$F(3, 13.35) = 0.60, p = 0.62$] or FICA [$F(3, 13.17) = 1.03, p = 0.40$]. However, PSM significantly improved from baseline to post-infusion 5 [$F(3, 14.43) = 3.90, p = 0.03$]. We observed a trend toward a moderate negative correlation between baseline LSWM and MADRS ($r = -0.45, p = 0.13$). A moderate negative correlation was observed between LSWM and CRP ($r = -0.37, p = 0.23$), although this association was not statistically significant. Other domains, including FC, did not show significant associations with either MADRS or CRP.

Conclusions: These preliminary results suggest that ketamine may improve fluid cognition independently of improvements in depression among individuals with mood disorders with mild to moderate symptoms of depression. The improvement was observed particularly in memory-related cognitive domains, which showed correlations with both depression severity and CRP. These findings should be replicated in larger samples and with the inclusion of additional inflammatory markers.

Keywords: IV- Ketamine, Cognition, CRP, Depression, Bipolar Disorder

Disclosure: Nothing to disclose.

P174. EYE-TRACKING AND EEG SIGNATURES OF SOCIAL INFERENCE DYSFUNCTION IN SCHIZOPHRENIA AND AUTISM SPECTRUM DISORDER

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Background: Deficits in social cognition are core features of schizophrenia (Sz) and autism spectrum disorder (ASD) and contribute substantially to functional disability. Impaired face emotion recognition (FER) disrupts the ability to interpret others' intentions and affective states, while abnormal visual scanning of social scenes reflects atypical saccade and fixation patterns that normally guide gaze toward socially salient features such as faces. In prior work, we used eye-tracking with The Awareness of Social Inference Test (TASIT) to examine comprehension of dynamic social interactions in Sz. TASIT presents short video vignettes in which actors engage in everyday exchanges, with the central character either sarcastic or lying. Compared with neurotypical (NT) controls, individuals with Sz made markedly fewer fixations to faces, and this aberrant pattern was accompanied by selective impairments in sarcasm detection, which depends strongly on facial cues. It remains unknown whether ASD shows similar abnormalities in visual scanning and fixation-related neural responses, and whether these reflect convergent or distinct alterations in how gaze and neural activity support comprehension of dynamic social interactions. Here, we combined eye-

tracking and EEG during TASIT to examine both visual scanning and fixation-related potentials (FRPs). FRPs are transient cortical responses elicited at fixation onset, well characterized in reading and scene viewing but rarely applied to clinical populations during naturalistic social inference. This multimodal approach enables identification of shared and disorder-specific alterations in gaze behavior and fixation-related neural processing during social information processing.

Methods: Participants were 18 NT, 32 Sz, and 18 ASD individuals. During TASIT, eye movements were recorded with a remote eye-tracking system (EyeLink). Fixation locations and saccades were quantified offline. To assess scanning variability, we calculated the z-transformed distance of each participant's gaze position relative to the NT mean on a frame-by-frame basis, yielding distributions of divergence values compared across groups and conditions. Fixations were also analyzed with respect to frame-level features including faces, motion, and luminance. EEG was recorded continuously with a 64-channel active electrode system (BrainVision). FRPs were derived by time-locking the EEG signal to fixation onset, and mean amplitude and peak latency were obtained in the 80–120 ms post-fixation window over occipital sites. Single-trial time-frequency analyses were performed using adaptive complex Morlet wavelets (3–12 cycles) over a 2.5-s window. From these decompositions, inter-trial coherence (ITC) and total power were computed in the 2–40 Hz range with 0.3 Hz resolution.

Results: Both ASD and Sz groups were significantly impaired on TASIT comprehension relative to NT controls ($F = 9.3, p < 0.001$). In Sz, deficits were most pronounced for sarcasm detection ($F = 5.5, p = 0.025$). Eye-tracking revealed abnormal scanning in both clinical groups, with 33.2% (Sz) and 40.4% (ASD) of fixations diverging more than 1.5 standard deviations from the NT mean. FRP amplitude was significantly reduced in Sz ($F = 5.1, p = 0.029$) and ASD ($F = 17.8, p < .001$). In ASD, FRP peak latency was markedly delayed ($F = 44.6, p < 0.001$), and greater delay correlated with poorer sarcasm detection ($r = -0.60, p = 0.008$). Frequency-domain analyses showed robust theta/alpha activity (5–10 Hz) at fixation onset in NT participants, which was significantly reduced in both clinical groups ($p < .01$). An ANCOVA controlling for group membership confirmed that sarcasm detection was positively associated with power in the FRP latency window ($F = 4.8, p = 0.033$). Finally, an exploratory deep-learning classifier integrating time- and frequency-domain FRP features achieved >90% accuracy in distinguishing ASD from NT and >80% in distinguishing Sz from both NT and ASD.

Conclusions: This is the first study to integrate eye-tracking and EEG to characterize shared and disorder-specific alterations in Sz and ASD during naturalistic social cognition. Both clinical groups exhibited fewer fixations to faces and diminished fixation-related neural responses, suggesting a common disruption in processing socially salient features. In contrast, delayed FRP latency in ASD indicates a disorder-specific alteration in the timing of neural responses during social inference. Together, these findings demonstrate the value of combining behavioral and neural measures to capture dynamic aspects of social information processing and support the development of translational biomarkers of social cognitive dysfunction across neuropsychiatric disorders.

Keywords: schizophrenia, ASD, social cognition, EEG, eye-tracking

Disclosure: Nothing to disclose.

P175. A COMPARISON OF A MULTIVARIATE NEURAL PATTERN VERSUS AMYGDALA RESPONSE FOR REGULATION OF NEGATIVE EMOTION

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Background: Emotion regulation is a core psychological function that can be accomplished using numerous strategies. One of the most common strategies taught in cognitive-behavioral therapy is cognitive reappraisal, which involves thinking about a situation differently to alter feelings. A picture induced negative emotion signature (PINES) has been shown to reflect negative emotion with greater accuracy than individual brain regions (e.g., the amygdala), but it remains unclear whether this measure is sensitive to emotion regulation.

Methods: In a sample of 82 young adults, we examined brain activation using fMRI while participants completed an emotion regulation task. The task presented participants with neutral or negative images with instructions to passively observe the images or cognitively reappraise them to feel less negatively. After each image, participants self-reported how negatively they felt. We applied PINES to the fMRI data, and also extracted activation levels from the left and right amygdala during negative images with reappraisal or passive observation, and to neutral images. We tested the effect of condition using a linear mixed-effects model.

Results: While amygdala activation did not differ between the cognitive reappraisal and passive observation conditions during presentation of negative imagery, PINES level was reduced during the cognitive reappraisal condition ($d = 0.27$, $p = 0.02$). During the reappraisal condition, decreased PINES level was associated with lower self-reported negative affect ($\rho = 0.27$, $p = 0.02$) but amygdala activation was not significantly associated with negative affect.

Conclusions: In our study, individuals instructed to use cognitive reappraisal reduced negative emotion neural pattern activation but not amygdala activation. This suggests that cognitive reappraisal engages a broader set of brain regions to down-regulate negative affect.

Keywords: Emotional regulation, Amygdala, Cognition

Disclosure: Nothing to disclose.

P176. NEUROCOMPUTATIONAL MARKERS OF GOALS AND HABITS OVER THE COURSE OF INPATIENT TREATMENT IN OPIOID ADDICTION

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Background: Heroin and other opiate overdose-related deaths continue to devastate our communities, necessitating exploration of neuroscience-based addiction severity and recovery biomarkers. A potential basic-science informed target is the motivational control over drug use that shifts as addiction progresses from goal-directed [model-based, via ventromedial prefrontal cortex (vmPFC) and ventral striatum] to habitual (model-free, via dorsal striatum), for which there exists scarce evidence in human studies.

Methods: Responding to the need for alternative, naturalistic efforts and addressing a longstanding translational question in addiction research, we estimated motivational control using parametric logistic regression and computational reinforcement learning models applied to the two-step decision task, administered via smartphone over eight weeks to 17 inpatients with opioid use disorder (OUD) and 18 healthy controls (HC). We distinguished reward-guided decision making in the two-step decision task as model-free (i.e., repeating decisions based solely on reward history) and model-based (i.e., repeating decisions based on a more sophisticated model of task structure). To reveal potential goal-directed behavior and brain relationships, participants also underwent a baseline fMRI scan. Here, we estimated the neural processing of drug and nondrug cues, as well as the regulation of the former via reappraisal (as compared to the savoring of nondrug food cues)—a PFC (e.g., anterior cingulate, a node of the salience network) mediated strategy to reduce drug cue reactivity.

Results: Consistent with previous studies that reported lower model-based choice in alcohol use disorder as associated with relapse, the logistic regression on stay probabilities as a function of previous choice revealed only the model-free strategy in OUD (Beta = 0.37, SE = 0.14, Z = 2.56, $p < 0.011$). In contrast, the HC exhibited a hybrid strategy, with both model-based (Beta = 0.28, SE = 0.09, Z = 3.00, $p = 0.003$) and model-free (Beta = 0.71, SE = 0.13, Z = 5.55, $p < 0.001$) influences evident, consistent with patterns in the general population. Computational models of choice strategies similarly indicated a trend for lower recruitment of model-based decision making (indexed by the goal-directed weighting parameter w) in OUD vs. HC, $t(32.42) = 2.01$, $p = 0.053$. Controlling for demographics, lower model-based choice in OUD was associated with higher naturalistic cue-induced craving (i.e., in response to a salient drug-themed movie, $F(1,12) = 5.14$, $p = 0.043$). Higher model-based choice was associated with higher vmPFC activity during drug vs. nondrug cue processing in OUD [$F(1,12) = 5.92$, $p = 0.031$], suggesting that the recruitment in this key region related to goal-directed control may be conferring resilience in the face of salient drug cues. Furthermore, higher model-based choice was associated with lower anterior cingulate engagement during drug cue reappraisal [$F(1,12) = 7.67$, $p = 0.012$], suggesting that in OUD exhibiting better goal-directed control, less recruitment of this hub of the salience network implicated in emotion regulation may be required to attenuate drug cue reactivity.

Conclusions: Using a cutting-edge approach that intersects computational, naturalistic and longitudinal methods, we leverage goal-directed control as a sensitive marker of craving and the neural correlates underlying drug cue reactivity and its regulation. These findings underscore a promising target that could inform motivation-based, translational, and neuropharmacological treatment avenues to tackle the ongoing opioid epidemic.

Keywords: opioid use disorder, Habitual decision-making, goal-directed control, cue-reactivity, Functional MRI (fMRI)

Disclosure: Nothing to disclose.

P177. ECOLOGICAL MOMENTARY ASSESSMENT OF MULTIPLE AFFECTIVE DIMENSIONS AND CRAVING FOR RELAPSE PREVENTION IN OPIOID USE DISORDER

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Background: Reuse and relapse remain the modal outcomes in opioid use disorder (OUD), even among individuals receiving gold-standard treatment. This highlights the need to better understand the predictors of relapse, to enable better interventions. An emerging area of research has emphasized real-time assessment of affect and craving, which are known to impact decision-making and opioid use, and are thus being considered as candidate targets for novel just-in-time interventions. However, previous studies have typically focused on either isolated emotions (e.g., anger) or potentially overly broad affective categories (i.e., negative affect), which may not fully capture the affective landscape of people with OUD. Moreover, the extent to which craving and other affective experiences represent separable dimensions contributing to reuse vulnerability and resilience remains unclear, particularly at the within-person level. To address these gaps, here we employed exploratory factor analysis (EFA) on densely sampled ecological momentary assessment (EMA) data, allowing us to reduce multiple self-reported affective states into a smaller set of latent affective factors. We then evaluated how these factors differed in their expression and temporal dynamics between individuals with and without OUD, and how they related to addiction-relevant cognitive decision-making processes.

Methods: Participants were treatment-engaged OUD participants ($N = 87$, 30 female) and comparison controls ($N = 77$, 34 female) enrolled in a longitudinal smartphone-enabled EMA study probing 26 affective states in real-time, three times daily, for 28 days. Measures included traditional “positive” items (e.g., in a good mood, feeling connected) and “negative” items (e.g. feeling sad, feeling the need to escape) as well as urge-related items (e.g., urge to do something risky/impulsive, or use opioids). Once daily, participants were also prompted to complete two brief cognitive tasks, assessing risky decision-making and impulsive choice, respectively. The affective data was tested for suitability to be analyzed with EFA using Spearman’s correlations of each of the 26 items, application of the Kaiser-Meyer-Olkin test, and Bartlett’s test of sphericity, all of which were passed. Factor number determination was guided by eigenvalues, scree plots, and parallel analysis, which suggested solutions ranging from 3 to 8 factors, with a 4-factor solution providing the best balance of parsimony and explanatory power. The resultant 4-factor structure was robust to analytic variations and contained no between-factor correlations of greater than 0.7, supporting factor separability. Factor means and variances were compared between the OUD and control groups, as well as those who did and did not return to opioid use over the study period. Daily factor scores were also tested for their relationship to cognitive task behavior.

Results: The application of EFA identified four dimensions in the data: “High Arousal Negative Affect”, “Low Arousal Negative Affect”, “Positive Affect”, and “Craving and Negative Self-Perception”. Wilcoxon rank-sum tests revealed that control and OUD groups differed significantly in their expression of all four factors, with the OUD group having higher mean factor scores over the study period for “High” and “Low Arousal Negative Affect”, as well as “Craving and Negative Self-Perception”, and lower mean scores for “Positive Affect” relative to controls (all $z_{162} > |3.15|$, $p < 0.002$). With the exception of “Positive Affect”, the OUD group also demonstrated a higher degree of within-person variability of each factor over time (standard deviation of factor scores) than controls ($z_{162} > |3.03|$, $p < 0.003$). Furthermore, within participants with OUD, those who used opioids during the study period had significantly higher mean factor scores for “High” and “Low Arousal Negative Affect”, as well as “Craving and Negative Self-Perception”, but not “Positive Affect”, compared with those who maintained abstinence ($z_{85} > |2.52|$, $p < 0.02$). Those who used also reported experiencing more variability in “High Arousal Negative Affect” and “Craving and Negative Self-Perception” over time ($z_{85} > 2.07$, $p < 0.04$). Analyses linking within-person changes

in daily factor scores to cognitive task behavior and contextual cues associated with opioid reuse are ongoing.

Conclusions: These findings suggest a richer, multidimensional affective experience in OUD than is typically captured by valence-based models. Moreover, the identified dimensions were differentially expressed in the OUD group, discriminated participants who returned to opioid use during the study, and varied at least partly independently over time. This work provides a more nuanced framework to characterize affective contributions to relapse and inform the development of targeted, adjunctive interventions for OUD.

Keywords: computational psychiatry, opioid dependence, decision-making, affect

Disclosure: Nothing to disclose.

P178. INVESTIGATING DENTATE GYRUS ACTIVITY IMPAIRMENTS IN OLD AGE AND THEIR IMPACT ON COGNITIVE DECLINE

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Background: Aging is associated with a progressive decline in memory, primarily affecting episodic memory, which involves the storage and retrieval of personal experiences linked to specific contexts and emotions. These deficits are associated with impairments in contextual discrimination, the process by which similar experiences are encoded as distinct representations to prevent interference and false memories. The dentate gyrus of the hippocampus plays a central role in contextual discrimination by sparsely activating distinct neuronal ensembles. Remarkably, the dentate gyrus retains the ability to generate new granule cells throughout life. Adult-born granule cells enforce sparse coding and facilitate the spatial and rate remapping of mature granule cells (mGCs), thereby facilitating discrimination between similar contexts.

With aging, neurogenesis declines sharply, dentate gyrus activity becomes less sparse, and contextual discrimination deficits emerge. Yet, the precise network mechanisms driving these impairments, and whether they involve altered remapping dynamics, remain unknown.

Methods: To address this question, we combined 2-photon and 1-photon in vivo calcium imaging approaches to monitor mGCs activity in young adult (3-month-old) and aged (18- to 21-month-old) Dock10 transgenic C57BL/6J male and female mice ($n = 3-6$ for each sex/experimental group). Recordings were carried out in two complementary behavioral paradigms. In the first, head-fixed mice performed locomotion on a treadmill with diverse spatial and olfactory cues, allowing the characterization of both place cells and cue-responsive cells. In the second, freely moving mice underwent a contextual fear discrimination task involving two highly similar contexts: context A, which was paired with a mild foot shock, and context B, which was not paired with a shock. Successful discrimination was defined behaviorally as the ability of mice to selectively express conditioned freezing in context A but not B. Across both paradigms, we tracked the activity of mGC ensembles across days, thereby assessing their stability, remapping dynamics, and coding strategies in young versus aged networks. All analyses were carried out using the software GraphPad Prism 10. Data (mean $\pm$ SEM) were analyzed using Student t-test (two-tailed, paired or unpaired when appropriate) or one-way and two-way (repeated-measures when appropriate) ANOVA, then followed by Tukey’s comparison test when necessary. Statistical significance * $P \leq 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Results: We successfully recorded mGCs in both young and aged mice for both 2-photon and 1-photon experimental set up. Freely moving aged mice displayed lower contextual fear discrimination abilities than young animals. Our findings indicate that aged animals exhibit highly stable mGC ensemble activity, suggesting a hyperstability that constrains the formation of distinct representations. Cue cells were earlier to emerge in aged mice but were less well-tuned. At later time points (1wk), aged mice showed stronger and more specific cue responses. Furthermore, the balance between place coding and cue coding was disrupted, with aged networks showing an overrepresentation of stable cue responses. This cue bias may underlie age-related failures in discriminating overlapping contexts, highlighting a shift in the pattern separation strategy of the dentate gyrus with aging.

Conclusions: These results suggest that age-related hyperstability of mGC ensembles and the accompanying shift toward cue-dominated encoding contribute to deficits in contextual discrimination. Interventions aiming at enhancing adult neurogenesis may represent a promising strategy to rescue age-related decline in pattern separation abilities. Such findings may lead to clinical trials aimed at improving pattern separation in aged populations that display this deficit, including individuals with mild cognitive impairment or Alzheimer's disease.

Keywords: Brain-aging, In vivo Imaging, dentate gyrus, pattern separation

Disclosure: Nothing to disclose.

P179. A CORTICOSTRIATAL LEARNING MECHANISM LINKING EXCESS STRIATAL DOPAMINE AND AUDITORY HALLUCINATIONS

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Background: Hallucinations are false percepts frequently experienced by people with psychotic disorders. An established aspect of the neurobiology of hallucinations and other psychotic symptoms is elevated striatal dopamine, with pro-dopaminergic agents worsening and anti-dopaminergic drugs improving this symptom via striatal dopamine blockade. Interestingly, dopamine elevation in psychosis predominates in dorsal striatum. Some previous theories indeed centered around the functional role of dopamine excess in psychosis, although knowledge about dopamine heterogeneity has since soared. Specifically, in contrast with the well-established role of ventral striatal dopamine in reward learning, dopamine in certain parts of the dorsal striatum (including the so-called auditory striatum) responds to sensory features irrespective of reward. Anatomical evidence of specialized corticostriatal loops involving association sensory cortex and sensory dorsal striatum in primates similarly supports a role in perception and hallucinations. A plausible mechanistic account of hallucinations should thus admit such anatomo-functional heterogeneity, specifying the involvement of distinct striatal modules in hallucination generation.

Methods: Reanalysis of mouse data: As described in detail in the original work (Schmack et al., 2021), on each trial water-restricted mice reported their auditory percept (either a 1 kHz tone or white noise) by pressing a lever. Tone trials were presented on 50% of trials. Correct decisions were rewarded after a random delay period. The willingness to wait for reward has been previously validated as a measure of statistical decision confidence. Dopamine signals were recorded from both auditory

and ventral striatum using fiber photometry (GRAB-DA sensor) in $n = 6$ mice. In a separate set of mice ($n = 7$), dopamine terminals in the auditory striatum were chronically stimulated via a laser across blocks of approximately fifty trials, interleaved with baseline blocks in which the laser was turned off.

New experiments and analysis of human data: Core features of the human task were matched to the mouse task. White noise was constantly played in the background and on some trials, a pure 1 kHz tone was presented. To further separate the influence of sensory and reward contributions to choice, participants also reported how certain they were that a tone was present (i.e., signal confidence) on a visual analog scale. At the end of each trial, participants were told whether the tone was indeed present or absent and whether their choice resulted in an additional reward. All participants ($n = 290$) completed the Cardiff Anomalous Perception Scale (CAPS).

Results: Mouse data: In the fiber photometry data, we found that the amplitude of signal-evoked model auditory striatal dopamine, but not ventral striatal dopamine, was modulated by the previous trial's stimulus identity (auditory, 95% CI $[-0.087, -0.017]$, $p = 0.0030$; ventral, 95% CI $[-0.18, 0.022]$, $p = 0.15$). In the optogenetic stimulation data, we found that false alarms, hits, signal confidence all exhibited a significant increase from baseline as a function of blocks within a stimulation session (false alarms across session, 95% CI $[0.22, 0.71]$, $p < 5 \times 10^{-4}$; hits, 95% CI $[0.024, 0.41]$, $p = 0.028$; signal confidence, 95% CI $[0.0051, 0.037]$, $p = 0.010$). These analyses suggest that dopamine dynamics in the mouse auditory striatum but not ventral striatum resembles sensory prediction error, and that dopamine-mediated plasticity mechanisms in the auditory striatum play a role in learning sensory expectation.

Human data: Leveraging our design to isolate stimulus-history effects, we found that hallucination propensity (CAPS) score was positively associated a tendency to report hearing a signal (including on noise trials, i.e., false alarms) following signal trials (Previous stimulus $\times$ CAPS: 95% CI $[0.037, 0.11]$, $p < 5 \times 10^{-4}$). Critically, this differential effect of stimulus history on choice was mirrored by a similar significant effect on signal confidence following signal trials (Previous stimulus $\times$ CAPS: 95% CI $[0.017, 0.044]$, $p < 5 \times 10^{-4}$), in contrast to the effect of reward history (Previous reward $\times$ CAPS: 95% CI $[-0.017, 0.0082]$, $p = 0.45$). These results suggest that biased hallucination-like percepts are consistent with selective alterations in sensory learning and sensory corticostriatal plasticity due to elevated sensory prediction error signaling.

Conclusions: Dopamine dynamics in the mouse auditory striatum mirrored sensory prediction errors essential for learning sensory expectation, and dopamine stimulation in this region gradually biased perceptual beliefs indicating a plasticity-based mechanism. In an auditory detection task, human behavior showed both sensory and reward learning, but only changes in the former correlated with hallucination proneness and additionally captured behaviors associated with clinical hallucinations. A single perturbation in a biologically constrained circuit model – enhanced dopaminergic sensory prediction errors in dorsal sensory striatum – is sufficient to explain hallucinations through a local change in corticostriatal plasticity that biases internal beliefs about sensory states. These findings reveal a potential computational circuit mechanism underpinning the link between excess striatal dopamine and auditory hallucinations, and provide a unifying account of influential hallucination-related results across paradigms and species.

Keywords: Computational Neuroscience, Auditory hallucinations, Dopamine, Corticostriatal circuit

Disclosure: Nothing to disclose.

P180. DISTINCT ROLES OF AFFECT-DRIVEN AND NON-AFFECTIVE IMPULSIVITY IN EXPLORE-EXPLOIT DECISION-MAKING AND REWARD VALUE ENCODING IN DEPRESSION

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Background: Depression is characterized by a constellation of symptoms across mood, cognition, motivation, and behavior, making it one of the most heterogeneous, prevalent, and disabling psychiatric conditions worldwide (Lee 2023, Ernst 2020). Among factors contributing to this heterogeneity is impulsivity, a multifaceted construct reflecting the balance between long-term goals and immediate urges that is strongly associated with suicidal thoughts and behaviors (Bruno 2023, Checn 2015, Whiteside 2001). Impulsivity can be affect-driven, involving rash actions triggered by heightened emotional states, or non-affective, reflecting a stable, context-independent tendency toward hasty decision-making. Evidence suggests these subtypes differentially influence outcomes in depression (Bruno 2023), yet the mechanisms underlying these distinctions remain unclear.

Disruptions in reward-guided decision-making and neural value encoding are well-documented in depression but vary widely across individuals (Chen 2015, Brown 2020). Explore-exploit decision-making tasks provide a powerful framework for quantifying these processes by measuring how individuals balance sampling uncertain options against exploiting known rewards. However, little is known about how distinct forms of impulsivity shape explore-exploit behavior and associated representations of reward value in depression. This study addresses this gap by examining relationships between affect-driven and non-affective impulsivity, explore-exploit decision-making, and value encoding to clarify mechanisms contributing to depression's heterogeneity.

Methods: We recruited 94 older individuals with depression (ages 49–80 y; $M = 61.7y$, $SD = 6.1$; 53 females) from the Pittsburgh community and assessed trait impulsivity using the UPPS-P scale (5), which yields four facets: two affect-driven (negative and positive urgency) and two non-affective (lack of perseverance and lack of premeditation). During fMRI, participants completed an explore-exploit decision-making task in a one-dimensional continuous space, deciding when to stop a revolving green within a 5-s interval to earn rewards (Moustafa 2008). Probabilistic feedback was determined by two conditions with independently varying reward probability and magnitude, producing expected values that increased or decreased over time.

We used multilevel modeling in R to capture two model-free behavioral metrics: response shifts after feedback (exploration) and convergence on optimal responses (exploitation). Trial wise expected values were derived using the SCEPTIC selective maintenance reinforcement learning model (Hallquist 2019) and Matlab VBA toolbox (Daunizeau 2014). fMRI images were pre-processed, and signals from ventral prefrontal cortex (vPFC) subregions and subcortical areas (striatum, hippocampus, and amygdala) were deconvolved using a leading hemodynamic deconvolution algorithm to estimate stimulus-locked neural activity (Dombrovski 2020). Multilevel models tested associations between impulsivity facets, model-derived expected values, and BOLD responses, controlling for age and sex, with significance thresholded at $pFDR < 0.05$.

Results: Positive urgency was linked to increased response shifts regardless of feedback ($\chi^2 = 15.7$, $p < 0.001$), whereas non-affective impulsivity was associated with greater shifts specifically after reward omission (lack of perseverance: $\chi^2 = 6.0$, $p = 0.01$; trend for lack of premeditation: $\chi^2 = 3.1$,

$p = 0.08$). Lack of perseverance also predicted convergence on the best response time, a measure of exploitation, ($\chi^2 = 17.0$, $p < 0.001$), while no other impulsivity facets significant related to exploitation.

Both negative and positive urgency were associated with diminished maximum value encoding across all vPFC subregions during all task phases (offline, online, and post-feedback). In subcortical regions, affect-driven impulsivity was associated with blunted value encoding in control network-connected striatal regions both offline and post-feedback ($pFDR < 0.05$). In contrast, lack of perseverance was associated with enhanced value encoding in the vPFC control subregion (online and post-feedback) and hippocampus (post-feedback, $pFDR < 0.05$), with lack of premeditation showing a similar but nonsignificant pattern.

Conclusions: These findings reveal distinct neural and behavioral mechanisms linking affect-driven and non-affective impulsivity to decision-making processes in depression. Affect-driven impulsivity was associated with heightened exploratory behavior and broadly blunted value encoding across the vPFC and striatum, suggesting reduced integration of reward information. In contrast, non-affective impulsivity was selectively related to exploration following negative feedback, reduced exploitation, and enhanced value representations in regions of the vPFC and striatum connected to the control-network that are less commonly engaged during value processing in nonpsychiatric populations (Papale 2025). This pattern may reflect compensatory overreliance on higher-order cognitive systems during decision-making, potentially driven by cognitive overload. Together, these results highlight heterogeneous pathways through which different forms of impulsivity may shape maladaptive thinking and behavior in depression and underscore the importance of distinguishing impulsivity subtypes to inform targeted interventions.

Keywords: explore-exploit dilemma, impulsivity, Depression subtypes, computational psychiatry

Disclosure: Nothing to disclose.

P181. NAO (NEUROSCIENCE AND AI-OPTIMIZED PLATFORM): A MULTI-MODAL COMPUTATIONAL PLATFORM FOR INFORMING TARGET AND INDICATION SELECTION, BIOMARKER READOUT, AND PATIENT STRATIFICATION IN CNS DRUG DEVELOPMENT

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Background: Neural drug development has a high failure rate. There is a critical need for data-driven insights starting from selecting the right drug targets and indications, to developing and validating biomarkers, and finally for clinical trial enrichment. The core goal is to map neural mechanisms causing harmful circuit dysfunctions to quantifiable phenotypes in order to effectively develop therapies that are guided by precision measures. The NAO (Neuroscience and AI Optimized) platform is developed and tested to address a critical unmet need: indication positioning, endpoint measure selection, and quantitative patient selection - all informed by human neuroimaging signals.

Methods: The NAO platform is designed to enable neuroimaging-informed biomarker development for neural disorders. Here we showcase NAO for informing neural drug R and D with a neurophenotype mapping use case in psychosis spectrum disorders using two independent samples from the B-SNIP consortium. We analyzed brain-behavior relationships using two

independent samples from the B-SNIP consortium. Neurophenotype mapping results were computed in discovery (BSNIP-1, $n = 393$), replication (BSNIP-2, $n = 298$), and combined datasets ($n = 691$). Behavioral decomposition by nonnegative matrix (NMF) factorization was performed on combined cognitive (BACS) and psychopathology symptoms (PANSS) to derive a low-rank symptom solution. Next, we used a functional neural parcellation to reduce dimensionality of the neural data and increase signal to noise. Mass univariate regression was used to map symptom scores across all subjects to global brain connectivity (GBC) from resting-state functional magnetic resonance imaging, creating group-level neurophenotype maps for each of the NMF factors and composite scores. Split-half replication (100 runs) was performed for each NMF component both for the full brain map and for the top 25% of symptom-relevant neural parcels.

Results: Key features of NAO include: (i) automated and scalable processing of large datasets with extensible processing modules; (ii) a data lake comprising gene expression brain maps as well as aggregated neuroimaging and phenotyping data from thousands of subjects across indications; (iii) multi-modal neuroimaging preprocessing and feature extraction, including both human-engineered and AI-derived features; (iv) algorithms for linking mechanism to clinical presentation; (v) interactive and customizable report generation for delivering analytic insights, enabling researchers to evaluate datasets and make decisions on drug development processes. Behavioral decomposition using NMF yields an interpretable and replicable solution (cosine similarity = 0.75 to 0.995 across the five components between Discovery and Replication cohorts). Neurophenotype associations between NMF scores and GBC vary in strength and replicability across the five behavioral components. R-values for split-half replicability for each NMF are as follows (whole brain|top 25%): NMF1 (0.24|0.59), NMF2 (0.08|0.15), NMF3 (0.40|0.81), NMF4 (0.53|0.92), NMF5 (0.11|0.29). NMF4 showed a higher level of replicability in the combined dataset than either discovery or replication alone, suggesting that cohort effects may exist. In contrast NMF2 showed consistent low split-half replicability, suggesting that covariation for this symptom dimension may not be suitably captured by the present approach. Additional analyses evaluating effects only in patients with severe symptoms (total PANSS score ≥ 60) and in patients with non-zero NMF scores in each component showed effects that were similar to the main results (min $r = 0.54$, mean $r = 0.80$). Sex and age did not differ significantly between discovery and replication. Brain-behavioral statistics remain fairly strong for most NMF components (Z score range of -5.75 to 5.54). A proof-of-concept analysis restricting the split-half analysis to the top 25% of parcels improved the replicability of the brain-behavior map, particularly for NMF4. This suggests that restricting brain-behavior maps to symptom-relevant circuits may improve replicability of results. Some behavioral components may associate more strongly with neural metrics beyond GBC; exploration is underway.

Conclusions: This work opens the possibility of developing new clinical scales optimized to represent variation in key neural circuits that can be targeted therapeutically, helping to advance the development of accessible precision medicines. The NAO platform presents an informed approach to CNS drug development that aligns with the future of personalized medicine and enables researchers to more quickly develop more effective therapeutic solutions. The NAO platform can support drug R and D by enabling and accelerating access to insights from a growing number of multi-modal datasets.

Keywords: Computational Cognitive Neuroscience, Computational Modelling of Neuroimaging, neuroinformatics, Psychosis spectrum symptoms

Disclosure: Nothing to disclose.

P182. PROBABILISTIC COGNITIVE STATE MODELING (PCSM) REVEALS ALTERED NEURAL DYNAMICS OF SERIAL AND PARALLEL INFORMATION PROCESSING ASSOCIATED WITH CALLOUS-UNEMOTIONAL TRAITS

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Background: Callous-unemotional (CU) traits describe a severe antisocial phenotype characterized by persistent criminal behavior, blunted empathy, and poor treatment response. These traits are associated with impairments in information processing. Influential bottleneck models (e.g., Baskin-Sommers, A., and Brazil, I. A. (2022)) propose that individuals with elevated CU traits over-rely on serial processing modes (i.e., sequential, rigid processing strategies) and have difficulty transitioning to parallel processing modes (i.e., simultaneous information processing strategies) for flexible integration of affective and cognitive information streams. Such bottlenecks have associations with reduced emotional response, lack of remorse, and persistent antisocial behavior. Identifying the cognitive and neural dynamics that drive these impairments is critical for developing effective interventions.

Yet most existing studies rely on static paradigms and analytic techniques that average across trials or embed static assumptions on processing modes. A key feature of information processing is the dynamic transitions between strategies to adapt to context and cognitive capacities. Static assumptions on designs and analyses limit insight into moment-to-moment neural dynamics and how processing strategies adapt (or fail to adapt) across contexts. Moreover, it is well known that serial and parallel processing are involved in cognitive processes such as cognitive control and working memory, yet the traditional neuroimaging contrasts—using specialized static designs intended to induce serial and parallel processing—cannot capture whether CU traits are linked to rigid persistence in one mode or inefficient transitions between modes during core cognitive functions.

To overcome this, I empirically validated and applied Probabilistic Cognitive State Modeling (PCSM), a novel framework that uses Gaussian mixture hidden Markov modeling (GMM-HMM) of trial-wise fMRI to derive metrics of dynamic processing. PCSM leverages learned state transition dynamics to compute Dsp (a temporal index of processing persistence vs. flexibility) and Bnode (regional contributions to these tendencies), providing a richer window into neural mechanisms of CU traits.

Methods: PCSM was applied to a subset of ABCD data (ages 9–10; balanced by sex; selected for variation in CU traits). Trial-wise finite-impulse-response models estimated fMRI responses during the stop signal task. Multivariate regional time series were fit with a GMM-HMM (empirically validated number of k states = 3 and mixtures = 2). From the learned state and transition dynamics, PCSM metrics were derived: Dsp (a temporal index of information-processing across the entire brain) and Bnode (the spatial node-level contributions). Mixed-effects models tested associations with CU traits, adjusting for age, sex, site, motion, SES, and IQ.

Results: The GMM-HMM yielded stable transition structure across cross-validation folds, supporting reliable estimation of PCSM metrics. Higher CU traits were associated with more time in serial processing and reduced switching as indicated by higher Dsp ($\beta = 1.25$, $p < 0.001$) and attenuated Bnode weights in dorsolateral prefrontal ($\beta = -0.85$, $p = 0.002$) and temporoparietal ($\beta = -0.61$, $p = 0.021$) regions. Effects remained after covariate adjustment and were not explained by general cognitive ability.

Conclusions: PCSM provides the first dynamic evidence that CU traits are linked to inflexible neural state dynamics and reduced recruitment of control and social-cognitive regions—patterns

consistent with bottleneck accounts of impaired affect–cognition integration. By moving beyond static contrasts, PCSM fills a key methodological gap, providing a framework to quantify how processing strategies unfold in real-time. By applying computational properties of PCSM, these findings advance mechanistic understanding of CU traits, opening new avenues for precision treatment in youth with antisocial tendencies.

Keywords: BOLD fMRI signal, Callous-unemotional traits, Computational Methods

Disclosure: Nothing to disclose.

P183. MULTIMODAL CONNECTOME-WIDE MARKERS OF DISRUPTIVE BEHAVIOR IN YOUTH: INSIGHTS INTO SEX-SPECIFIC NEURODEVELOPMENTAL PATHWAYS

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Background: Disruptions in large-scale networks supporting the cognitive control of emotion—including frontoparietal, default mode, and salience networks—are implicated in disruptive behavior disorders in youth. Most studies examining the connectome, or brain-wide connectivity, and disruptive behavior in children are cross-sectional. Thus, it is unclear whether baseline patterns of brain-wide connectivity (functional or structural) predict later changes in disruptive behavior severity. While sex differences in network connectivity have been observed developmentally, it is largely unknown whether males and females show distinct connectomic markers of disruptive behavior. Addressing these gaps is crucial for clarifying neurodevelopmental circuit mechanisms underlying child mental health and identifying robust brain biomarkers for targeted interventions. This study investigates if multimodal imaging markers, from resting-state functional and diffusion weighted imaging-derived structural connectomes, predict cross-sectional and prospective changes in disruptive behavior in youth. We also examine if sex-specific predictive networks might index divergent disruptive behavior trajectories for males versus females.

Methods: This study included a transdiagnostic sample of 3,630 youth from the Adolescent Brain and Cognitive Development (ABCD) Study. Children were 9–10 years at baseline and 11–12 years at 2-year follow-up ($n = 1830$ males; $n = 1800$ females). Functional connectomes were derived from resting-state with eyes open. Structural connectomes were derived from quantitative anisotropy (QA), which measures white matter integrity. The Child Behavior Checklist (CBCL) Externalizing Behavior Problems subscale was used as a continuous measure of disruptive behavior. Connectome-based Predictive modeling—a machine learning, data-driven approach with 10-fold cross-validation—identified networks predicting disruptive behavior. Permutation testing (1000 iterations) estimated significance. Separate models were conducted for functional and structural connectomes. First, cross-sectional brain-behavior associations were tested in the total sample and separately for males and females. Next, for prospective prediction, we investigated whether baseline connectomes predicted changes in disruptive behavior (i.e., baseline to 2-year follow-up) using a CBCL raw change score. All models accounted for age, cognition/IQ, and motion. Prospective models also accounted for baseline disruptive behavior levels. FDR-correction was applied to permutation p -values.

Results: Cross-sectionally, functional ($p = 0.07$, $RMSE = 5.8$, $pFDR = 0.009$) and structural ($p = 0.05$, $RMSE = 5.6$, $pFDR = 0.006$) connectivity predicted disruptive behavior; positive and negative

edges contributed comparably. Sex-stratified analyses revealed significant predictions only in males: for functional connectivity ($p = 0.06$, $RMSE = 6.4$, $pFDR = 0.03$) this was driven by negative edges (decreased connectivity), and for structural connectivity ($p = 0.07$, $RMSE = 6.2$, $pFDR = 0.01$) this was driven by positive edges (increased connectivity). Neither modality was significant in females (all $pFDR > 0.2$).

Prospectively, only the male subgroup showed significant prediction of symptom change. Increased functional connectivity at baseline predicted increases in symptom severity ($p = 0.07$, $RMSE = 4.9$, $pFDR = 0.03$). In contrast, decreased structural connectivity at baseline predicted increases in symptom severity ($p = 0.06$, $RMSE = 4.8$, $pFDR = 0.006$). No prospective associations reached significance in females (all $pFDR > 0.40$).

Shared patterns of partially overlapping hub nodes were identified across functional and structural connectivity in cognitive control (medial frontal, frontoparietal), social functioning (default mode, salience), and sensorimotor networks. In males, the direction of brain-behavior association was distinct across modalities: decreased functional but increased structural connectivity predicted greater baseline symptoms, while increased functional but decreased structural connectivity predicted worsening symptoms. Distinct patterns of network connectivity predicted symptoms cross-sectionally with a distributed pattern across cognitive control systems, and prospectively with a focal pattern spanning salience, medial frontal, and subcortical networks.

Conclusions: Both functional and structural connectomes provide distinct and complementary insights into disruptive behavior severity in childhood. Male-specific neural markers suggest divergent neurodevelopmental pathways for disruptive behavior in boys versus girls. A multimodal, network-level framework holds promise for refining clinical phenotyping and guiding circuit-based targeted treatments for childhood disruptive behavior disorders.

Keywords: Multimodal imaging, disruptive behavior disorders, connectomics, sex differences, cognitive control network

Disclosure: Nothing to disclose.

P184. NEURAL REPRESENTATIONS SUPPORTING ABSTRACTION AND GENERALIZATION UNDER CONTINUAL LEARNING IN HUMANS

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Background: Abstraction and generalization are essential for adaptive behavior in novel situations, and dysfunction in these processes contributes to neuropsychiatric disease. Recent work in humans and monkeys has shown how the brain may encode an abstract representation of the environment in the representational geometry—or relationships between neural activity patterns associated with specific task conditions—of single-neuron activity. However, these observations are typically made after learning has converged, leaving open the question of how humans acquire and deploy abstract knowledge, and how the underlying neural representations form. Answering this question is essential for understanding individual variation in abstract learning and generalization, particularly the pathological deviations observed in neuropsychiatric illness.

Methods: To address this question, we introduce a novel computational model of abstract learning and generalization and apply it to a novel behavioral task that human participants perform during fMRI. The model, which builds on the successor

representation, disentangles the contribution of successive levels of abstraction: from stimulus-response associations to generalizable task schema. We fit the model to the behavior of healthy human participants ($N = 39$; 31 female) as they learn the correct response and outcome contingencies for a set of stimuli ("local associative learning"). Unbeknownst to participants, the contingencies depend on two latent contexts that alternate in blocks of trials, which participants could exploit to infer the correct response to all stimuli after a change in just one stimulus ("cross-context learning"). Across sessions, novel stimuli are used, allowing participants to transfer abstracted knowledge (i.e., a schema) of the task structure to these new task instances ("generalization"). When fit to behavior, the model quantifies—for each participant, on each trial—how much each level of learning is contributing to the newly acquired abstract knowledge, thereby predicting a corresponding change in the representational geometry that reflects this new knowledge. We test these predictions in fMRI using standard univariate regression to relate the magnitude of the predicted change in knowledge to the magnitude of the BOLD activity on every trial—thereby probing the dynamics of the neural representations on the timescale of learning.

Results: Human participants successfully learned and exploited the task's latent structure, inferring the correct response on the first encounter with a stimulus after a context change (above-chance accuracy: $t(38) = 7.7$, $p = 1.2 \times 10^{-9}$). Crucially, participants learned continually across multiple timescales throughout the task: gradual improvement within a block of stable contingencies; increasingly rapid one-shot updating (i.e., inference) when contingencies switched between blocks; and gradual zero-shot learning (i.e., generalization) across sessions as they transferred abstracted knowledge to novel task instances. Our model captured the dynamics of human learning at all of these timescales. Moreover, the model revealed that participants relied on all levels of learning throughout the task, with a gradual evolution from lower to higher levels, ultimately relying heavily on generalization to map novel stimuli to familiar abstracted states. Probing the neural correlates of these levels of learning confirmed their continual and independent roles across a network of regions implicated in relational learning. In particular, BOLD activity in hippocampus, amygdala, and orbitofrontal cortex correlated more with learning attributed to generalization than to the other levels of abstraction (cluster extent, $p < 0.05$; Gaussian random field correction for familywise error rate). Moreover, our model predicted an increasing role for entorhinal cortex—a putative source of abstracted, schema-like knowledge. Indeed, entorhinal BOLD activity was increasingly correlated with generalization-based learning. Finally, the model related individual variation in neural activity to the tendency to abstract general structure: the more a participant's BOLD activity was attributed to generalization-based learning, the greater their behavioral performance on key inference trials that required abstract knowledge.

Conclusions: We combine a novel computational model, human behavior, and fMRI to uncover the trial-to-trial process by which humans gradually learn abstract knowledge and the dynamics by which the neural signals representing this knowledge form. We show that humans learn continually over multiple levels of abstraction, with increasing reliance on more abstract levels, including generalization. Consistent with these predictions, we find a distributed network of brain regions encoding different levels of abstract learning, with an increasing role for hippocampus and entorhinal cortex in supporting generalization. Finally, individual variation in the neural signals associated with generalization predicts behavioral performance that depends on inferential reasoning. Our approach offers a computational framework for probing the dynamics of the neural representations underlying abstract learning, both at the timescale of individual trials and the level of individual participants. This framework ultimately may

help uncover the pathological variation in abstraction and generalization underlying neuropsychiatric disease.

Keywords: learning generalization, Computational Cognitive Neuroscience, Computational Reinforcement Learning Model, Functional MRI (fMRI), abstraction

Disclosure: Nothing to disclose.

P185. MAPPING ENHANCER-GENE LINKS IN DORSOLATERAL PREFRONTAL CORTEX ACROSS THE LIFESPAN IDENTIFIES PUTATIVE REGULATORY MECHANISMS UNDERLYING PSYCHIATRIC DISORDERS

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Background: Therapeutic targets supported by human genetic data are more than twice as likely to advance successfully to approved drugs. Genetic studies have been highly successful in psychiatry, with genome-wide association studies (GWAS) implicating hundreds of risk loci. However, most of GWAS variants lie in non-coding regions, making it challenging to identify causal genes that could be targeted therapeutically. Multimodal single-cell technologies, which jointly measure chromatin accessibility and gene expression, now allow high-resolution, cell-type-specific mapping of how genetic risk variants influence gene regulation. In this study, we constructed enhancer-gene regulatory maps in postmortem dorsolateral prefrontal cortex (DLPFC) samples across the human lifespan. Our approach allowed us to link genetic risk to specific genes and pathways, providing new insights into disease mechanisms and prioritizing novel targets for therapeutic development.

Methods: We integrated seven single-nucleus multiome datasets (paired snRNA-seq and snATAC-seq) from neurotypical DLPFC across developmental timepoints from early fetal to late adulthood ($n = 95$ samples; $>700,000$ cells). Using SCENT, a non-parametric bootstrapping method, we inferred enhancer-gene links in six broad cell types: excitatory neurons, inhibitory neurons, astrocytes, oligodendrocytes, oligodendrocyte precursor cells, and microglia. We intersected our enhancer-gene maps with fine-mapped GWAS loci for bipolar disorder (BIP, 298 loci), depression (MDD, 697 loci), and schizophrenia (SCZ, 287 loci).

Results: Preliminary results from one dataset ($n = 20$ samples; 125,991 cells; 539,703 million enhancer-gene pairs) identified 16,629 significant enhancer-gene links ($FDR < 0.10$), reflecting cell-type-specific regulatory relationships. Integration with fine-mapped SNPs ($PIP > 0.1$) revealed candidate regulatory mechanisms for 3 BIP, 7 SCZ, and 4 MDD GWAS loci. Our putative enhancer target genes were strongly enriched for genes previously implicated in neurodevelopmental disorders via loss-of-function variants (enrichment ratio = 3.55, 95% CI 3.22–3.89, $p = 3.8 \times 10^{-218}$). For previously prioritized genes, we refined putative mechanisms by newly implicating specific DLPFC cell types. For example, for BIP we linked FAM135B to a SNP (rs4909802) located in a putative inhibitory neuron-specific enhancer. At previously unresolved non-coding loci, we identified likely causal variants within putative neuron-specific enhancers for CRTAC1 (rs61873668, MDD) and UBR5 (rs7835270, SCZ). Analyses in the full dataset and validation using Hi-C and allele-specific expression analyses are underway.

Conclusions: Our enhancer-gene maps highlight how common regulatory variation and rare coding mutations may converge on shared neurodevelopmental pathways to confer psychiatric risk, and further demonstrate the power of multimodal single-cell

approaches to resolve non-coding GWAS loci. By linking fine-mapped variants to high-confidence target genes in specific cell types, our framework enables systematic prioritization of therapeutic candidates. Once validated, future efforts will focus on refining and validating these targets, which represent tractable and biologically grounded opportunities for drug development in psychiatry.

Keywords: ATAC-seq, single-cell, RNA-seq, Genomic, integration of GWAS with brain omics

Disclosure: Nothing to disclose.

P186. ELUCIDATING THE COGNITIVE PROCESSES THAT UNDERLIE INHIBITORY CONTROL DEFICITS IN COCAINE USE DISORDER: TRAIT IMPULSIVITY AND PAST MONTH COCAINE USE PREDICT INEFFICIENT PROCESSING

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Background: Inhibitory control captures the ability to inhibit a prepotent response, such as approaching a reward cue. Deficits in inhibitory control are frequently found in substance use disorders, including cocaine use disorder. This cognitive processing bias is thought to be of clinical relevance, since such deficits could make abstinence or substance use reduction challenging. Importantly though, studies that assess inhibitory control in clinical populations typically quantify deficits using summary statistics of task behavior, such as rate of inhibitory failures. An emerging body of work shows that such indices suffer from poor test-retest reliability, undermining their utility in research that seeks to link dimensions of psychopathology with cognitive processes.

Hierarchical and computational modeling can address these statistical concerns. Further, computational models provide insight into the generative processes that produce a behavioral bias. Drift diffusion models (DDMs) are a particularly promising class of computational models: A growing literature implicates low drift rate – a marker of inefficient processing of a goal-relevant stimulus – in externalizing psychopathology. Here, we applied hierarchical drift diffusion models to characterize the decision processes that underlie inhibitory control. To understand their role in cocaine use disorder, we then linked these decision processes with impulsivity and recent cocaine use.

Methods: Participants were 136 adults with moderate-to-severe cocaine use disorder. Participants were recruited into a randomized controlled trial utilizing contingency management to understand the biopsychosocial health benefits of reduced cocaine use. This study focused on data obtained during the baseline visit, prior to the start of the intervention.

During the baseline visit, participants completed a Cued Go/No Go Task, commonly used to measure inhibitory control. In this task, participants are instructed to respond by pressing a button to one stimulus ('go' target) and to withhold a response to the other stimulus ('no go' target). Prior to viewing the target stimulus, participants saw a visual cue ('go' or 'no go' cue), which was probabilistically related to the target stimulus. Participants had up to 1000 ms to respond to the 'go' stimulus. Participants completed 250 trials of the task. Trial-level reaction time and accuracy data from this task were fit using hierarchical drift diffusion modeling. After verifying model convergence, maximum a posteriori (MAP) estimates were extracted from the posterior distribution and were used in analyses linking traits with DDM MAP estimates.

At the baseline visit, participants also completed the Barratt Impulsiveness Scale (BIS). Our study uses the BIS total score as a measure of trait impulsivity. Participants self-reported on cocaine

use over the past 30 days. To test whether DDM parameters were systematically related to impulsivity or cocaine use, we regressed MAP estimates onto BIS total scores and onto recent cocaine use.

Results: Drift rate was higher for 'Go' trials (MAP_Go = 5.91, [5.66, 6.18]; MAP_NoGo = 3.97, [3.61, 4.37]), especially when the cue was congruent with the target stimulus (MAP_GoXCongruent = 6.15, [5.81, 6.49]). Trait impulsivity, as measured with the BIS total score, predicted lower drift rate to the 'Go' target, both on congruent ($B = -0.27$, $p = 0.002$) and incongruent ($B = -0.22$, $p = 0.009$) trials. This effect held when controlling for age, sex, and race (p 's < 0.020). Similarly, greater cocaine use within the past 30 days predicted lower drift rate to the 'Go' target on congruent trials ($B = -0.22$, $p = 0.011$), and this effect held when controlling for age, sex, and race ($B = -0.22$, $p = 0.013$).

Conclusions: Among individuals with moderate-to-severe cocaine use disorder, greater BIS total scores and past month cocaine use predicted lower drift rate on 'Go' trials. Our results align with studies linking inefficient evidence accumulation with externalizing psychopathology and substance use. Altogether, our findings highlight one lower-level cognitive process that contributes to poor inhibitory control among individuals with cocaine use disorder.

Keywords: Inhibitory control, cocaine use disorder, impulsivity, drift diffusion model

Disclosure: Nothing to disclose.

P187. DIFFERENTIAL VULNERABILITY OF STIMULUS-LOCKED AND PERSISTENT GAMMA OSCILLATIONS TO SCHIZOPHRENIA-ASSOCIATED SYNAPTIC PERTURBATIONS: A COMPUTATIONAL MODELING STUDY

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Background: Core cognitive deficits in schizophrenia (SZ) include impairments in visuospatial working memory (vsWM). Different aspects of vsWM rely on γ -oscillations with distinct dynamics: stimulus-locked γ -oscillations are essential for stimulus encoding, while persistent γ -oscillations maintain information in the absence of stimuli. Our prior computational studies showed that multiple synaptic alterations found in the PFC of SZ synergistically (> the sum of individual impact) reduce the power of γ -oscillations. In this study, we investigated whether this synergistic effect of synaptic alterations extends to both stimulus-locked and persistent γ -oscillations, and whether these two types of γ -oscillations exhibit distinct vulnerabilities to such alterations.

Methods: We constructed a population model network of a local cortical microcircuit composed of pyramidal neurons (PNs) and parvalbumin-positive interneurons (PVIs), capable of generating either stimulus-locked or persistent γ -oscillations. The two oscillatory regimes were implemented as separate network configurations, differing in the level of NMDA conductance among PNs. We then simulated three synaptic alterations previously reported in the PFC of SZ: (1) reduced strength of excitatory inputs onto PVIs from PNs, (2) reduced strength of inhibitory inputs from PVIs to PNs, and (3) increased variability in the strength of excitatory drive across PVIs. Oscillation power was quantified for each regime using power spectral density analysis. Synergistic effects were defined as reductions in γ -power under combined perturbations that exceeded the linear sum of individual effects.

Results: The model robustly generated either stimulus-locked or persistent γ -oscillations (peak ~40 Hz), depending on the NMDA conductance among PNs. All three synaptic alterations associated with SZ reduced γ -power individually. When combined, these

alterations produced a greater-than-additive (synergistic) reduction of γ -power in both regimes, and this effect was mainly driven by lower excitatory synaptic strength to PVIs. Finally, the magnitude of this synergy was greater for persistent γ -oscillations than for stimulus-locked γ -oscillations, indicating that persistent dynamics are more vulnerable to converging synaptic perturbations.

Conclusions: Our findings suggest that persistent γ -oscillations exhibit greater vulnerability to the synergistic effects of SZ-associated synaptic alterations than stimulus-locked γ -oscillations. Given that stimulus-locked γ -oscillations arise in early sensory areas while persistent γ -oscillations emerge primarily from PFC, this differential vulnerability implies that a greater magnitude of synaptic alterations may be required to disrupt γ -oscillations in early sensory areas than in the PFC in SZ. These results also highlight the potential value of region-specific, dose-sensitive therapeutic strategies that account for the distinct circuit-level resilience of sensory versus prefrontal γ -oscillations.

Keywords: Schizophrenia (SZ), gamma oscillations, computational modeling

Disclosure: Nothing to disclose.

P188. TARGETING FRONTOSTRIATAL REWARD SYSTEM CONNECTIVITY AND ANHEDONIA IN YOUTH DEPRESSION USING CONTINUOUS THETA BURST STIMULATION PLUS POSITIVE AFFECT TRAINING: INDIVIDUAL DIFFERENCES AND NETWORK PRECISION ARE KEY

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Background: Anhedonia involves disrupted positive affect (PA), is a hallmark of depression, and has been linked to hyperconnectivity between the dorsomedial prefrontal cortex (dmPFC) and nucleus accumbens (NAcc). In previous work, a single session of continuous theta burst stimulation (cTBS) to dmPFC decreased NAcc-dmPFC functional connectivity (FC), with corresponding improvement in PA. We investigated whether 2-week cTBS combined with PA training is associated with alleviated anhedonia and depression, reduced NAcc-dmPFC FC, and correlation between FC change and anhedonia change.

Methods: 50 youth (15–25 years) with current depression received twice-daily cTBS over 10 days targeting a personalized dmPFC location, with PA training between sessions. Resting-state fMRI and symptom severity pre/post TBS + PA were collected. Analyses addressed change in symptoms and NAcc-dmPFC FC, and their association. The actual target network (ATN) was calculated by estimating whole-brain connectivity of individualized electrical fields, and mutual information between NAcc FC and ATN FC was computed as an index of targeting accuracy.

Results: 42% of participants responded to cTBS+PA. Depression [$F(1,44.5) = 37.3$, $p < 0.001$, semi-partial $R^2 = 0.26$], anhedonia [$F(1,43.7) = 9.4$, $p < 0.005$, semi-partial $R^2 = 0.05$], and anxiety [$F(1,43.7) = 12.2$, $p < 0.005$, semi-partial $R^2 = 0.05$] improved with TBS, and overall NAcc-dmPFC FC remained unchanged [$F(1,45.0) = 0.1$, $p = 0.76218$, semi-partial $R^2 = 0.00$]. Participants with greater decrease in FC experienced more improvement in anhedonia [$B = 0.42$ (SD = 0.16), $\beta = 0.34$, $t = 2.7$, $p < 0.05$]. Moreover, those with greater overlap between reward network and ATN showed greater decrease in FC [$B = 2.51$ (SD = 1.23), $\beta = 0.32$, $t = 2.0$, $p < 0.05$].

Conclusions: cTBS+PA shows promise as an intervention for anhedonic depression in youth, with individual differences in FC change relevant to clinical outcomes. Precise network targeting could be critical for modulating the intended network and reducing symptom severity.

Keywords: depression, anhedonia, cTBS, ventral striatum, dmPFC

Disclosure: Nothing to disclose.

P189. CULTURED CONNECTIONS: BRIDGING CLINICAL BIOMARKERS AND BRAIN TISSUE MODELS IN TREATMENT-RESISTANT DEPRESSION

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Background: Bench-to-bedside research leads to focused biomedical advances but can be hampered by cost and gaps between patient experience and model systems. In vitro models of patient-derived tissue, as opposed to animal or generic cell-line models, offer great promise to fill this void towards personalized biomarker development at scale. In an ongoing study of glutamatergic signaling in the fast-acting antidepressant effects of ketamine in treatment-resistant depression (TRD), we generated cerebral organoids from individual patients for mechanistic and predictive assays. We report preliminary data towards the synaptic hypothesis of ketamine action, and intriguing involvement of cell developmental and blood-brain barrier pathways.

Methods: Patients aged 18–70 are being recruited for TRD study NCT03973268 (NIH Clinical Center, Bethesda, MD, USA). After medication taper, they are randomized to receive the AMPAR antagonist perampanel (8mg p.o.) or placebo (1:2 ratio) prior to ketamine (0.5 mg/kg i.v.). Montgomery-Asberg Depression Rating Scale (MADRS) is scored serially to test AMPARs as necessary for fast-acting antidepressant action of ketamine. Surrogate plasticity measures including magnetoencephalography (MEG) and blood samples are obtained. Peripheral mononuclear cells are reprogrammed to induced pluripotent stem cells (iPSCs). After embryoid body formation, cerebral organoids are generated using STEMdiff protocols (StemCell Technologies). Organoids from TRD participants across multiple protocols were developed to 60 days, compared to those from psychiatrically healthy controls (HC), and challenged with (2R,6R)-hydroxynorketamine (HNK) in vitro prior to bulk RNA sequencing. RNAseq was analyzed with Ingenuity Pathway Analysis (IPA, QIAGEN). Pathway analyses are reported with ($-\log[p\text{-value}]$). Other statistical analyses were performed using SPSS version 28.0 (IBM). Benjamini-Hochberg p -values are shown for pathway analyses to adjust for type I errors.

Results: This interim analysis includes $N = 15$ patients. Baseline MADRS score was 28.0 ± 3.7 (SD). There were $n = 3/15$ responders by 4h after the first blinded drug and ketamine dosing, and $n = 4/15$ responders after a second blinded dosing. $N = 11$ individuals consented to cell line development. In comparison with HC ($n = 5$ TRD, $n = 6$ HC), expression pathways in TRD were downregulated ($-\log[p\text{-value}]$), including CREB signaling in neurons ($17.9[1.37\text{e-}15]$), phagosome formation ($17.0[5.65\text{e-}15]$), and S100 family signaling ($14.85[3.93\text{e-}13]$). Biological themes included immune response, bone differentiation, and cell proliferation. Following HNK application, these 3 expression pathways were significantly upregulated in the TRD cell lines ($3.5\text{--}5.4[0.04\text{--}3.03\text{e-}3]$), but not in the HC lines. Examining blood-brain barrier pathways, neurovascular coupling signaling (defined by 232 markers) was downregulated in TRD vs HC (BH $p = 7.3 \times 10^{-5}$), with downregulated

KCNJ2, PTGS1, HSP70, and PLC gamma, and upregulated GRM1, P2RX1, ADORA2A, and AQP8. These expression alterations were reversed following HNK for all but AQP8 in TRD ($p \leq 1 \times 10^{-4}$), but not HC organoids.

Conclusions: This preliminary analysis supports multiple synaptic and nonsynaptic pathway deficits involved in TRD, which may be reversed by a putative fast-acting antidepressant. The ongoing study remains blinded but will characterize in vitro and in vivo responses to ketamine, HNK, and perampanel in parallel. The use of patient-derived cerebral organoids in the context of biomarker studies and clinical trials may vastly amplify the usefulness of valuable patient time and painstakingly collected deep clinical phenotyping.

Keywords: Brain organoids, Induced pluripotent stem cells (iPSCs), Depression, AMPA receptors, (2R,6R)-hydroxynorketamine

Disclosure: Nothing to disclose.

P190. NEURONAL EXOSOMES IDENTIFIED A BRAIN MITOCHONDRIAL PATHWAY OF EPIGENETIC PROGRAMMING OF TREATMENT RESISTANT DEPRESSION

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Background: Treatment resistant depression (TRD) is a leading cause of disability worldwide; there is a dearth of new mechanistic models for the development of better therapeutic strategies. Our group provided the first evidence that boosting mitochondrial metabolism of acetyl-L-carnitine (LAC) leads to a rapid and long-lasting antidepressant response in multiple rodent models by activating epigenetic processes of histone acetylation. Our translational work showed decreased levels of this pivotal mitochondrial metabolite LAC in clinical phenotypes of depression linked to childhood trauma and insulin resistance. Unlike the role of systemic mitochondrial metabolism, molecular mechanisms of brain mitochondrial metabolism and the related epigenetic pathways in TRD remain to be determined.

Methods: Here, we isolated neuronal exosomes from plasma of healthy control subjects and subjects with TRD in the context of transcranial magnetic stimulation (TMS). Next, we characterized key molecular markers of mitochondrial metabolism and the related pathways.

Results: Ongoing analyses showed a link between a key mitochondrial marker assayed in neuronal exosomes and a treatment resistant course of illness ($n = 93$, $p < 0.05$). These findings were specific to neuronal exosomes as no association was found in total (not neuronally-enriched) exosomes ($p = 0.7$). These phenotypes are also characterized by corresponding changes in the levels of key epigenetic markers important for histone acetylation. Ongoing experiments are aimed at determining the role of this pathway in the responses to TMS.

Conclusions: In all, these findings suggest that mitochondrial metabolism has important effects on epigenetic processes in patients with TRD and point to a novel model of mitochondrial-related epigenetic regulation as a mechanistic target for the rapid regulation of plasticity.

Keywords: Mitochondria, Exosomes, Molecular mechanisms, Epigenetics

Disclosure: Nothing to disclose.

P191. BLOOD-BRAIN BARRIER FUNCTIONAL IMPROVEMENTS IN NEUROVASCULAR WATER EXCHANGE RATE AS A MODIFIABLE TARGET IN ACCELERATED TRANSCRANIAL MAGNETIC STIMULATION FOR DEPRESSION

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Background: Treatment-resistant depression (TRD) is a major cause of disability. Evidence suggests the cerebrovascular system, particularly the blood-brain barrier (BBB), plays a role in depression and may be affected by transcranial magnetic stimulation (TMS). Lasting BBB effects of TMS suggest a combination of neuronal and non-neuronal mechanisms in antidepressant responses. A novel MRI technique measuring neurovascular water exchange rate (kw), linked to aquaporin-4 (AQP-4) activity, offers a reproducible, non-invasive marker of BBB function. This may bridge the neuronal and non-neuronal elements within the neurovascular unit, allowing investigation of rapid antidepressant mechanisms previously overlooked.

Methods: Using diffusion-prepared arterial spin label (DP-ASL) MRI, kw was measured in 25 healthy controls (HC) and 71 TRD participants at baseline. TRD participants then received accelerated intermittent theta burst (iTBS) TMS targeting either left dorsolateral (DLPFC) or dorsomedial prefrontal cortex (DMPFC) in an ongoing trial (NCT04982757). TMS was delivered with the MagVenture system using neuronavigation, 10 sessions/day over 5 days. BBB function and Hamilton Depression Rating Score – 17 item (HAMD-17) scores were assessed pre-treatment and one week after the treatment course.

Results: Regional kw values were significantly different between HC and TRD brains in two regions: right thalamus ($F = 9.2$, $p = 0.003$) and left inferior temporal gyrus ($F = 9.2$, $p = 0.003$). The right thalamus kw was reduced in HC compared to TRD (85.3 min^{-1} [22.5] vs 103.2 [26.2], respectively) and the left inferior temporal gyrus kw was increased in HC compared to TRD (86.4 [30.7] min^{-1} vs 64.2 [27.3] min^{-1} , respectively). Baseline left globus pallidus BBB function predicted pre-treatment HAMD-17 scores ($t = -2.29$, $p = 0.025$).

Conclusions: Findings suggest TMS effects in TRD may involve BBB modulation, particularly in patients with impaired BBB function. Differences in regional BBB function patterns exist between HC and TRD populations, and likely reflect underlying patterns of neurovascular network activity. Left globus pallidus BBB function emerged as a predictor baseline depression severity, possibly illustrating upstream neurovascular changes and AQP-4 activity. These findings highlight the BBB as a promising target for novel antidepressant strategies and an underexplored engageable biologic endpoint.

Keywords: Blood-Brain-Barrier, Neurovascular, TMS, Major Depressive Disorder

Disclosure: LivaNova, Advisory Board, Self.

P192. SPERMIDINE ALLEVIATES DEPRESSION VIA CONTROL OF THE STRESS RESPONSE

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Background: Depression is a major global health burden with limited therapeutic efficacy of current treatments and frequent side effects. The cellular stress response involves tight regulation of polyamine metabolism and autophagy, both of which are disrupted in stress-related psychiatric disorders. Imbalanced polyamine levels, particularly spermidine, have been identified as hallmarks of stress exposure and major depressive disorder (MDD). Importantly, antidepressant response is associated with increased plasma spermidine, suggesting a mechanistic link between polyamines, autophagy, and treatment success.

Methods: We combined human cohorts and mouse models to examine polyamine-autophagy interactions in acute and chronic stress, depression, and treatment response. In humans, metabolomic and autophagy assays were conducted in acute stress ($n = 22$ bungee jump participants) and in multiple depression cohorts ($n = 200+$, both sexes). A double-blind, placebo-controlled clinical trial ($n = 79$; 35 drug-naïve depressed, 44 healthy; both sexes) tested 3-week spermidine supplementation. Parallel mechanistic studies were performed in mouse models of acute (ASDS) and chronic social defeat stress (CSDS; $n = 5-20$ /group; both sexes), including pharmacological manipulations of glucocorticoid receptor signaling.

Results: In humans and mice, acute stress altered polyamine metabolism and transiently increased autophagy, linking psychological stress to cellular homeostasis. By contrast, chronic stress and depression were associated with disrupted spermidine regulation and reduced autophagic activity. In a clinical cohort, only patients responding to antidepressant therapy showed an increase in spermidine levels, suggesting a connection between polyamine metabolism and treatment success. Spermidine supplementation restored autophagy and alleviated depressive-like behavior in stressed mice. Importantly, in a randomized, placebo-controlled trial, three weeks of spermidine intake improved depressive symptoms in drug-naïve patients and enhanced autophagy in blood cells.

Conclusions: Our translational work identifies dysregulated polyamine-autophagy signaling as a hallmark of stress and depression. Spermidine supplementation restores autophagic balance, alleviates depressive symptoms, and may serve as a safe, well-tolerated, and cost-effective novel treatment approach for depression. The polyamine-autophagy axis thus represents both a biomarker and a therapeutic target in stress-related psychiatric disorders.

Keywords: Acute and Chronic Stress, Depression, autophagy, Cellular metabolism, Homeostasis

Disclosure: SPDLife, Founder, Spouse/Partner.

P193. COMPARING SHARED AND UNSHARED TRANSCRIPTOMICS ACROSS REPRODUCTIVE MOOD DISORDERS

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Background: Reproductive-related mood disorders (RMDs), including premenstrual dysphoric disorder (PMDD), perimenopausal depression (PMD), and postpartum depression (PPD), are distinguished by their appearance during periods of reproductive hormone change. Nevertheless, the ovarian steroid fluctuations that trigger each RMD are distinct, and the degree to which there is co-morbidity within RMDs is unclear. Here we investigate whether intrinsic transcriptomic differences (that may predispose certain women to RMDs) are shared between PMDD, PMD, and

PPD, to better define whether their genetic risk and cellular mechanisms converge.

Methods: We utilized AmpliSeq RNA-seq to re-characterize female patient-derived lymphoblastoid cell lines (LCLs) ($N = 56$) that were previously cultured in ovarian steroid-free conditions for RMD-specific experiments. Three differential expression ($p_{\text{nominal}} < 0.05$) gene lists (for either PMDD, PMD, or PPD, and matched RMD-controls) were generated via edgeR, then compared between RMDs.

Results: While each RMD had a similar quantity of nominally significant differentially expressed genes (DEGs) compared to their respective control group, only four genes (MLF1, UBAC2, DNAJC1, ARHGAP5) were shared across all RMDs, and no FDR-significant genes overlapped between RMDs. Moreover, we found that RMDs exhibited robustly different overall transcriptomic patterns, with PMDD and PPD showing primarily downregulated differential expression, and PMD showing predominantly upregulated differential expression relative to controls. Therefore, although PMDD and PMD shared the greatest DEG overlap, >50% of those shared genes were regulated in opposite directions. While certain steroid-sensitive genes (e.g., SNAI1, GATA3) were significant for one or more RMDs, no nuclear steroid receptors were DEGs. Notably, many previously identified RMD-related transcripts (e.g., CXCL10, WWTR1, JARID2) were significantly replicated in one RMD or another (as described for PMD, PPD, and PMDD, respectively), but all remained unshared across RMDs.

Conclusions: Despite PMDD, PMD, and PPD each having ovarian steroid-mediated triggers for affective symptoms, these preliminary findings suggest that the risk for RMDs may be consequent of largely unshared intrinsic transcriptomic differences or genetic substrates unique to each RMD.

Keywords: postpartum depression, Perimenopausal Depression, premenstrual dysphoric disorder, Transcriptomics

Disclosure: Nothing to disclose.

P194. RAPID-ACTING ANTIDEPRESSANT POTENTIAL OF DSP-3456, A NOVEL MGLUR2/3 NEGATIVE ALLOSTERIC MODULATOR, WITH TRANSLATIONAL EEG BIOMARKERS FROM PRIMATES TO HUMANS

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Background: Conventional antidepressants such as SSRIs require several weeks to demonstrate efficacy in major depressive disorder (MDD), and approximately one-third of patients are treatment-resistant. Ketamine is highly effective in treatment-resistant depression (TRD) but produces psychotomimetic side effects.

Group II metabotropic glutamate receptors (mGluR2/3) negatively regulate glutamate release and are implicated in the pathophysiology of depression. mGluR2/3 antagonists such as MGS0039 and LY341495 have shown rapid antidepressant-like effects in rodents without ketamine-like adverse effects. However, their clinical efficacy in TRD patients remains unconfirmed. DSP-3456 is a novel mGluR2/3 negative allosteric modulator (NAM) with a unique chemical structure, currently under development for TRD. We evaluated its pharmacological profile, efficacy, safety, and translational potential across species, focusing on electroencephalogram (EEG) biomarkers and comparisons with ketamine. Safety, tolerability, and pharmacokinetics (PK) of DSP-3456 were also assessed in healthy human subjects.

Methods: DSP-3456 was evaluated in vitro and in vivo in rodents and non-human primates. Assessments included PK, behavioral tests (forced swim test [FST], prepulse inhibition [PPI], Y-maze test), glutamate biosensor analysis, synaptic potentiation,

c-Fos expression, and EEG power spectrum analysis. A phase 1, randomized, double-blind, placebo-controlled single ascending dose (SAD) study was conducted in healthy humans to assess safety, PK, and brain action via EEG changes. EEG analysis focused on gamma-band activity as a marker of glutamatergic modulation.

Results: DSP-3456 selectively inhibited mGluR2 ($IC_{50} = 14.8$ nM) and mGluR3 ($IC_{50} = 60.4$ nM), with no significant activity at other mGluR subtypes. Radioligand binding assays confirmed its NAM mechanism, showing no binding to the glutamate orthosteric site at mGluR2. Off-target screening revealed high selectivity across a broad panel of receptors, transporters, ion channels, and enzymes.

A single dose of DSP-3456 (1 mg/kg, p.o.) significantly reduced immobility time in the rat FST at both 1 and 24 hours post-dose, with efficacy comparable to ketamine (10 mg/kg, s.c.). Plasma concentrations of DSP-3456 (1 mg/kg, p.o.) were near the IC_{50} values for mGluR2 and mGluR3 at 1 hour but fell below these levels at 24 h, indicating the sustained effects despite reduced exposure. Unlike ketamine (10 mg/kg, s.c.), DSP-3456 (up to 10 mg/kg, p.o.) did not impair PPI or Y-maze performance in rats, indicating a favorable behavioral safety profile.

DSP-3456 (1 and 10 mg/kg, p.o.) increased c-fos expression in the mouse medial prefrontal cortex (mPFC), similar to ketamine (10 mg/kg, s.c.), but did not induce c-fos expression in the striatum, unlike ketamine, indicating region-specific activation relevant to antidepressant efficacy without psychotomimetic effects. DSP-3456 (1, 3 mg/kg, p.o.) and ketamine (10 mg/kg, s.c.) significantly increased extracellular glutamate levels in the rat mPFC at 1 h post-dose. At 24 h post-dose, both DSP-3456 (1, 10 mg/kg, p.o.) and ketamine (10 mg/kg, s.c.) significantly increased the frequency of excitatory postsynaptic currents (EPSCs) in response to 5-HT in the rat mPFC. This effect was primarily mediated by cortical-cortical synapses within the apical tuft, indicating long-term synaptic potentiation.

DSP-3456 (1, 3 mg/kg, p.o.) and ketamine (10 mg/kg, s.c.) significantly enhanced relative gamma power (30–80 Hz), indicating that DSP-3456 effectively penetrates the brain and modulates glutamatergic neurotransmission. This gamma enhancement may serve as a surrogate marker for increased glutamate release. DSP-3456 also dose-dependently increased low gamma-band power (30–55 Hz) and decreased delta-theta (0.5–8 Hz)/low gamma power ratio in rhesus monkeys at plasma concentrations approximating the IC_{50} values for mGluR2 and mGluR3. Similar gamma-band modulation was observed following ketamine administration in rhesus monkeys at clinically relevant antidepressant doses.

In the SAD study, no serious adverse events were reported. The most frequently observed adverse events with exposure-related incidence were vertigo, nausea, vomiting, and headache. Plasma exposure increased dose-dependently, with peak concentrations at approximately 2 h post-dose, indicating favorable absorption. Resting-state EEG analysis showed a trend toward increased low gamma power and a significant reduction in the delta-theta/low gamma power ratio compared to placebo, consistent with preclinical findings.

Conclusions: DSP-3456, a novel mGluR2/3 NAM, was found to be safe and well tolerated in humans. Preclinical studies demonstrated that DSP-3456 exerts rapid and sustained antidepressant-like effects mediated by activation of glutamatergic neurotransmission and enhancement of synaptic plasticity in the mPFC, comparable to ketamine but without ketamine-like side effects. Gamma-band modulation by DSP-3456—characterized by increased low gamma power and decreased delta-theta/low gamma power ratio—was consistently observed in rats, non-human primates, and humans, reflecting glutamatergic engagement and suggesting its potential as a translational biomarker of antidepressant efficacy. These findings support DSP-3456 as a promising candidate for further clinical development in the treatment of TRD.

Keywords: mGluR2/3 negative allosteric modulator, Rapid-Acting Antidepressant, Clinical trial, Electroencephalography (EEG), Ketamine

Disclosure: Nothing to disclose.

P195. MULTI-MODAL MODELING OF KETAMINE AND PSYCHEDELIC MECHANISMS IN TREATMENT-REFRACTORY DEPRESSION (TRD)

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Background: Rapid-acting antidepressants (RAADs), including ketamine and psilocybin, have transformed the landscape for treatment-resistant depression (TRD) therapeutics [1–3]. Despite different primary receptor targets, both ketamine and psilocybin produce rapid clinical responses (within hours to days) and elicit similar biological correlates of treatment response, including increased prefrontal cortical glutamate levels [4–7], increases in magnetoencephalographic (MEG) spontaneous signal diversity (i.e., entropy and Lempel-Ziv complexity) that correlate with subjective experience intensity [8], and mTORC1-sensitive changes in structural and functional plasticity [9]. These observations suggest that ketamine, psilocybin, and other putative RAADs may converge on a shared downstream program that governs rapid response. Here, we sought to identify those common mediators by integrating (i) transcriptomic signatures from patient-derived iPSC cortical neurons exposed to four RAADs, (ii) serial cerebrospinal-fluid (CSF) proteomics collected after ketamine infusion, and (iii) multi-modal phenotyping—whole-blood RNA-seq, plasma cytokines, and resting-state MEG—in patients with TRD and healthy volunteers.

Methods: Ten-week-old iPSC cortical neurons were generated from patients with TRD ($n = 5$) and controls ($n = 5$) who had consented for iPSC generation during a previous Institutional Review Board (IRB) approved study at the NIH Clinical Center (NCT02484456). Neurons were treated for 6 h and 24 h with ketamine or (2R,6R)-HNK (1 μ M each), psilocybin or LSD (10 μ M each), or 0.1% acetonitrile vehicle before bulk RNA-seq (NovaSeq X Plus, ~60 M reads/sample; alignment with STAR/FeatureCounts). Differentially expressed genes (DEGs) were identified with Dream/limma ($p < 0.05$). To determine iPSC neuron resemblance to human central nervous system (CNS) changes, iPSC DEGs were compared with longitudinal CSF proteomics (Olink Explore; 40 min–24 h; six time-points) obtained after 0.5 mg/kg IV ketamine in healthy volunteers ($n = 9$, 54 samples) [10]. Overlap significance (hypergeometric; directionality binomial) and network topology (STRING v12 + Cytoscape 3.10) identified conserved hub markers across RAADs. Whole-blood RNA-seq from a randomized, double-blind ketamine crossover study (NCT00088699; TRD $n = 16$, HC $n = 11$) was collected at baseline and 4 h post-infusion; responders were defined as $\geq 50\%$ MADRS reduction at 24 h. Ingenuity Pathway Analysis (IPA) and xCell deconvolution tested immune pathways and cell-type shifts. Plasma cytokines (Bio-Rad 27-plex; log-normalized) were also assayed at 0, 4, 24, 72 h. To focus on treatment effects (ketamine vs placebo), linear mixed-effects models (Kenward–Roger df) collapsed over post-treatment time-points linked cytokines to MADRS changes while adjusting for crossover design (infusion order), depression (at baseline and between infusions), age, biological sex, and body mass index

(BMI). Resting-state MEG (30–50 Hz gamma) was recorded at baseline and 6–9 h post-infusion. MEG power maps were Talairach-warped in AFNI [11], brain-masked (cerebellum excluded), and gamma power normalized to the root-mean-square of the six canonical 2–100 Hz bands. Linear mixed-effects models (AFNI 3dLMER) assessed cytokine, diagnosis, and session (baseline, post-placebo, post-ketamine) effects.

Results: Across the four putative RAADs, 3157 differentially expressed genes (DEGs) showed strong pair-wise transcriptomic similarity $r = 0.70\text{--}0.88$ (all $p\text{FDR} < 2e-16$). Intersecting 24-h iPSC-neuron signatures with ketamine CSF proteomics yielded a 114-protein “RAAD signature network” enriched for cytokine/osteoclast pathways, with IL-15 and MCP-1 as topological hubs. IL-15 was upregulated across ketamine (CSF-protein), LSD (iPSC-gene), and psilocybin (iPSC-gene). Clinically, baseline ketamine responders (6/16) showed down-regulated IL-15 signaling gene sets and up-regulated B-cell-receptor pathways (IPA), both of which reversed 4 h post-ketamine in responders. xCell deconvolution confirmed post-ketamine reductions in naïve, memory and class-switched B-cells (all $p\text{FDR} < 0.05$) and increased NKT fractions. Plasma IL-7 (primary B cell driver) strongly predicted higher baseline cortical gamma power, reaching brain wide significance (main effect $p\text{FDR} < 0.05$) and strongest in subcortical structures ($p\text{FDR} < 10^{-14}$). Post-ketamine this relationship inverted, with IL-7 levels being associated with broad reductions in MEG gamma power throughout most default-mode network regions ($p\text{CLC} < 0.05$). Of 20 cytokines detectable in our multiplex panel, MCP-1 was the most strongly associated with antidepressant non-response. Conversely, cytokine ratios mechanistically-associated with IL-15 signaling, robustly predicted antidepressant efficacy (MCP-1/IL-7 $p = 0.0003$; IL-4/IFN- γ $p = 2.39e-05$), implicating IL-7/IL-15 rebalancing as central to therapeutic response.

Conclusions: Multi-omic integration across iPSC neurons, CSF, and blood reveals a conserved IL-7/IL-15–MCP-1 axis linking peripheral immune balance to central circuit plasticity and rapid antidepressant efficacy of ketamine, psilocybin and next-generation RAADs. Plasma cytokines (and mechanistically informed ratios) track leading response biomarkers (MEG gamma) and strongly predict clinical response to ketamine, offering translational markers for patient stratification and rational immunomodulatory augmentation.

Keywords: Multi-omics, Racemic Ketamine and metabolites, Psychedelics, magnetoencephalography, immune mechanisms

Disclosure: Nothing to disclose.

P196. VALIDATION OF A CLINICAL PHENOTYPE IDENTIFYING THE FIRST ANTIDEPRESSANT TRIAL FOR A MAJOR DEPRESSIVE EPISODE USING REAL-WORLD ELECTRONIC HEALTH RECORD DATA

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Background: Depressive disorders have an 18.5% lifetime prevalence in the U.S., resulting in a 28.9-year quality adjusted life expectancy loss, and over \$210 billion in costs annually. Antidepressant medications are the evidence-based and guideline-recommended first-line treatment for Major Depressive Episodes. However, only a reported 25–37% of patients report depressive symptom remission after the first antidepressant medication trial for depression, which contributes to the risk for and clinical and societal burden of treatment-resistant depression. Precision medicine strategies matching patients to the antidepressant medication that is most likely to work for them or

predicting if the patient is unlikely to have an adequate response to an antidepressant medication have the potential to improve early depressive disorder treatment response, which would decrease the clinical and societal burden of depressive disorders. To facilitate this goal, it is critical to define a sensitive and specific phenotype of patients trialing an antidepressant medication for the treatment of a depressive episode for the first time. The electronic health record (EHR) is an ideal data source to develop such a phenotype as it is an almost universally implemented tool in clinical practice with extensive clinical data. To date, a validated, EHR-based phenotype of a first antidepressant medication trial for a major depressive episode is lacking; previous EHR-based attempts have not tried to differentiate between antidepressant medication trials or had low specificity or sensitivity for depressive disorders due to use of claims datasets and lack of routine depressive symptom collection. This study aimed to define and validate an EHR-based phenotype of a first antidepressant trial for a major depressive episode using real-world clinical data.

Methods: Retrospective cohort study of electronic health record data in a large, diverse, integrated, community-based health care system. Universal depressive symptom screening occurs in this health system yearly for individuals with a history of depressive disorders and at regular time intervals after a new depressive disorder diagnosis using the patient health questionnaire (PHQ-9). The PHQ-9 is validated for depression screening across racial/ethnic groups with a cut-off score of ≥ 10 having high sensitivity ($>88\%$) and specificity ($>88\%$) for a Major Depressive Episode. The study time period included 1/1/2011–12/31/2023; which represented the start of universal depressive symptom screening implementation in the EHR. Inclusion criteria for our phenotype of interest (Cases) included age ≥ 18 -years-old at the time of an antidepressant medication fill at an outpatient setting and a PHQ-9 score ≥ 10 in the 18 months prior to or two weeks after the fill; no prior antidepressant medication fills with an international classification of diseases (ICD) depressive disorder diagnosis within 18 months prior to fill; enrolled with drug coverage one year prior to fill and six months after fill, allowing for 31-day gap. Exclusion criteria included ≥ 3 ICD diagnoses (on different dates) of bipolar, schizophrenia, or schizoaffective disorders in the 2 years prior to incident antidepressant fill date. Control subjects met the same criteria but were not prescribed an antidepressant medication. The San Diego Approach to Variable Validation (SDAVV) was used to validate the phenotype. This process systematically allows investigators to identify the number of charts needed to review to reach desired performance measure goals. The chart review sample size was determined by setting the target performance goal for a successful validation to a one-sided α -level lower confidence bounds of 0.05 for the estimated population and PPV and NPV to be above the pre-specified critical threshold of 0.95. This study was reviewed and deemed exempt by the Kaiser Permanente Northern California Institutional Review Board.

Results: Overall, 299,919 subjects met the phenotype case criteria, and 265,966 subjects met the control criteria. Of these, 65% identified as female, 55% as White, 18% as Hispanic, 13% as Asian, and 8% as Black. $N = 200$ charts were reviewed ($n = 100$ case and $n = 100$ control subjects) to meet our target performance goals for a positive predictive value (PPV)/negative predictive value (NPV) = 0.95 with a critical lower bound of 0.907. For both our phenotypic cases and controls, 99 subjects met criteria on chart review and 1 did not meet criteria, resulting in a PPV and NPV = 0.99. This EHR-based phenotype identified a first antidepressant trial for a major depressive episode with a sensitivity of 99% and specificity of 99%.

Conclusions: Using the specified inclusion and exclusion data, we obtained a PPV and NPV above the critical threshold and lower bound with 95% confidence, suggesting the code used to identify an EHR-based clinical phenotype of a first antidepressant

medication trial for a major depressive episode performed well. These criteria can be used in other EHR-based studies to identify validated cohorts of patients trialing an antidepressant medication for the first time to treat a depressive disorder, and for future studies examining precision medicine strategies for antidepressant medication treatment of depressive disorders.

Keywords: Depression, Real World Data, validation, Antidepressants

Disclosure: Cytel, Advisory Board, Self.

P197. DIMENSION-SPECIFIC MODERATORS OF KETAMINE RESPONSE: EFFECTS OF DIAGNOSIS AND MEDICATION EXPOSURE ON COGNITIVE VS SOMATIC SYMPTOMS IN DEPRESSION

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Background: Ketamine demonstrates rapid antidepressant effects, yet most evaluations rely on Patient Health Questionnaire-9 (PHQ-9) total scores, which can obscure heterogeneity in symptom change across domains. Cognitive/affective and somatic symptoms may follow distinct trajectories with different predictors of response. Capturing these domain-specific patterns may improve mechanistic understanding and highlight clinically meaningful moderators. By moving beyond total scores to examine dimensional outcomes, we can better identify patient factors shaping treatment response and refine precision strategies. We therefore examined cognitive/affective (Factor 1) and somatic (Factor 2) dimensions to test whether diagnosis and medication exposures moderated ketamine response.

Methods: We analyzed an intention-to-treat cohort of 1799 adults (mean age 41.4 [SD 13.6]; 56% women) with treatment-resistant depression, including major depressive disorder (MDD; $n = 1611$) and bipolar depression (BD; $n = 188$), treated with IV ketamine at a large outpatient network. Most patients ($n = 1565$) adhered to a standard protocol of four infusions over 14 days, while 234 were treated off-protocol or discontinued early. Depressive symptoms were assessed by PHQ-9 at baseline (≤ 30 days pre-treatment), infusion visits #1 and #4, and at +3, +9, and +14 days post-treatment. At intake, patients self-reported comorbidities: anxiety ($n = 1,304$), obsessive compulsive disorder (OCD; $n = 222$), post-traumatic stress disorder (PTSD; $n = 502$), substance use disorder (SUD; $n = 180$), and suicidal ideation ($n = 1,024$). Psychotropic use included antidepressants ($n = 1117$), antipsychotics ($n = 298$), benzodiazepines (BZ; $n = 462$), non-benzodiazepine sedatives (NBAS; $n = 275$), mood stabilizers ($n = 321$), stimulants ($n = 393$), and other agents ($n = 239$). Exploratory factor analysis of PHQ-9 items identified Factor 1 (cognitive/affective) and Factor 2 (somatic). Symptom trajectories were modeled using linear mixed-effects regression with log-transformed time as the primary variable. Fixed effects included baseline severity, age, sex, diagnosis, comorbidities, and medication exposures. Participant intercepts were random. Secondary analyses examined moderators by sex and age (18–64 vs ≥ 65). False discovery rate correction controlled for multiple testing.

Results: Ketamine produced robust improvement in both domains. Factor 1 declined over time ($\beta = -0.25$, 95% CI -0.26 to -0.24 , $p < 0.001$), paralleled by Factor 2 improvement ($\beta = -0.23$, 95% CI -0.24 to -0.22 , $p < 0.001$).

Sex effects: At baseline, males reported slightly higher Factor 1 scores ($\beta = 0.09$, 95% CI 0.04 – 0.15 , $p = 0.003$); Factor 2 differences were nonsignificant ($\beta = 0.05$, 95% CI -0.01 – 0.10 , $p = 0.17$). Significant sex-by-time interactions were observed. For

Factor 1, sex differences emerged early and widened across follow-up (Day 1: $\beta = 0.13$, 95% CI 0.02 – 0.24 ; Day 45: $\beta = 0.24$, 95% CI 0.13 – 0.35). For Factor 2, interactions became evident by Day 10 and persisted (Day 10: $\beta = 0.11$, 95% CI 0.01 – 0.21 ; Day 45: $\beta = 0.15$, 95% CI 0.05 – 0.26).

Primary diagnosis: For Factor 1, BD and MDD trajectories converged (Time $\times$ BD $\beta = -0.00$, 95% CI -0.02 to 0.02 , $p = 0.998$). For Factor 2, BD patients improved faster (Time $\times$ BD $\beta = -0.03$, 95% CI -0.05 to -0.01 , $p = 0.004$).

Medication moderators: BZ blunted Factor 1 improvement ($\beta = 0.11$, 95% CI 0.04 – 0.18 , $p = 0.003$) with only a nonsignificant trend for Factor 2. NBAS were unrelated to Factor 1 ($\beta = 0.05$, 95% CI -0.03 – 0.13 , $p = 0.24$) but impaired Factor 2 ($\beta = 0.11$, 95% CI 0.04 – 0.18 , $p = 0.009$).

Comorbidities: OCD was associated with slower improvement on Factor 1 ($\beta = 0.14$, 95% CI 0.06 – 0.23 , $p = 0.003$) and Factor 2 ($\beta = 0.15$, 95% CI 0.07 – 0.23 , $p = 0.001$). PTSD predicted faster Factor 1 improvement ($\beta = -0.12$, 95% CI -0.18 to -0.05 , $p = 0.001$). SUD showed no significant moderation.

Conclusions: Ketamine produces robust antidepressant effects across both cognitive/affective and somatic domains, yet moderators influence these trajectories in dimension-specific ways. BZ attenuated cognitive/affective improvements, while NBAS impeded somatic recovery. Patients with BD and MDD showed comparable gains on cognitive outcomes but diverged on somatic symptoms, suggesting transdiagnostic mechanisms for affective relief but diagnosis-specific differences in somatic response. These findings highlight the limitation of total scores, which can obscure clinically meaningful variation, and underscore the value of domain-level monitoring to guide personalized treatment. Minimizing BZ use may help preserve ketamine's cognitive benefits, while reducing NBAS may optimize somatic recovery. In this large real-world cohort, moderators exerted distinct effects across symptom dimensions, reinforcing the importance of a dimensional framework for both mechanistic research and precision clinical care.

Keywords: Ketamine, Dimensional outcomes, Depression, Treatment moderators

Disclosure: Nothing to disclose.

P198. META-ANALYSIS OF NEURAL CORRELATES OF TREATMENT RESPONSE IN DEPRESSION

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Background: Major depressive disorder (MDD) is highly prevalent and debilitating, with a significant subset of patients failing to respond to frontline treatments. Encouragingly, several promising and relatively novel treatment modalities (e.g., ketamine) are increasingly offered alongside well-established approaches (e.g., SSRIs, electroconvulsive therapy, cognitive behavioral therapy), providing clinicians with additional options and new hope for treatment-resistant patients. However, clinicians have limited research evidence to draw on when making critical treatment choices, as findings informing individualized treatment selection remain sparse, and the precise neurophysiology underlying treatment response for any given approach is unclear. Seeking to fill these gaps, recent years have seen an explosion of studies investigating brain regions and networks that correlate with treatment response in depression – but, methods and results have greatly varied, highlighting the need for a meta-analytic approach that can quantitatively synthesize findings across studies and identify reliable patterns. The present coordinate-based meta-analysis tested findings from studies investigating neural

correlates of antidepressant treatment response, spanning a wide range of evidence-based modalities and functional imaging analysis approaches to identify the most reliably implicated brain regions, converging across treatment modalities and imaging analysis approaches.

Methods: A systematic search of major publication databases (PubMed, APA PsycInfo, MEDLINE, Web of Science) was conducted for studies published prior to 4/25/2024. Search terms were combinations of key imaging terms (e.g., “fMRI”, “network”), treatment terms (e.g., “medication”, “treatment”) and variations of the term “depression” through use of a wildcard. This resulted in over 10,000 articles, and all abstracts were subsequently screened for eligibility. Of these, 623 articles continued to full-text review, with 154 articles ultimately deemed fully eligible and included in analyses. Eligibility criteria required that participants experienced clinically significant unipolar depressive symptoms (e.g., MDD diagnosis) and received an evidence-based treatment for a therapeutic duration; and required that the study reported whole brain functional magnetic resonance imaging (fMRI) or positron emission tomography (PET) analyses predicting treatment response or testing changes over the course of treatment. Coordinates of reported effects were extracted, and multilevel kernel density analysis was conducted to identify brain regions that most consistently served as putative moderators (i.e., baseline predictors of subsequent treatment response) or mediators (i.e., regions showing change over the course of treatment). Preliminary analyses tested for global effects across moderator and mediator analyses to identify regions of the brain that are implicated in antidepressant treatment response. A Monte Carlo simulation with 10,000 iterations produced the familywise error rate threshold used to correct for multiple comparisons.

Results: A total of 154 studies were included in the meta-analysis, representing 5203 individual participants. Several regions were consistently implicated in antidepressant treatment response across studies, including areas of the basal ganglia and dorsomedial prefrontal cortex/dorsal anterior cingulate cortex. Additional preliminary results included the right lingual gyrus, left precuneus, and areas of orbitofrontal cortex.

Conclusions: The present study was a large meta-analysis seeking to identify regions of the brain implicated in antidepressant treatment response, sweeping across a wide array of evidence-based treatment modalities and imaging analysis approaches. Preliminary results point to brain areas associated with both depression risk and pathology, as well as putatively related cognitive processes such as reward learning. Immediate next steps for these analyses include (1) identifying brain regions that serve as moderators or mediators of treatment response, respectively and (2) investigating the extent to which brain systems underpinning responses to different treatments are shared or divergent. Advancing our understanding of the neural regions and networks central to antidepressant treatment response may ultimately improve the accuracy of treatment response prediction, aid in the optimization of treatment selection, and inform the development of novel therapeutic approaches (e.g., neuromodulation or neurofeedback targeting these circuits).

Keywords: Depression, meta-analysis, treatment

Disclosure: Nothing to disclose.

P199. MODULATION OF STRESS RESPONSE SIGNALING BY TAI CHI CHIH IN PROMOTING REMISSION TO REFRACTORY LATE-LIFE DEPRESSION

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Background: Geriatric depression presents unique clinical challenges, as it is often characterized by limited responsiveness to conventional treatments and high rates of treatment resistance. These complexities underscore the urgent need for innovative and integrative treatment approaches. Emerging evidence suggests that adjunctive mind-body interventions, such as of Tai Chi Chih (TCC), may serve as valuable complements to standard pharmacological and psychotherapeutic regimens.

Methods: Data were obtained during a 3-month randomized clinical trial (clinicaltrials.gov: NCT02460666) comparing TCC (N = 89; 69.7% Female; 82.0% Caucasian) versus Health Education and Wellness training (HEW; N = 89; 75.3% Female; 83.2% Caucasian) on mood and cognitive functioning in adults age ≥60 years with treatment-resistant depression (≥15 on the 24-item Hamilton Rating Scale for Depression, HAM-D, despite stable antidepressant treatment) and mild cognitive impairment. Participants were stable on antidepressant therapies for at least 4 months before starting the trial. TCC and HEW participants significantly improved on the HAM-D, Geriatric Depression Scale, and Connor-Davidson Resilience Scale at 3- and 6-months post-treatment ($p < 0.05$, no between group differences). Only TCC demonstrated significant improvement in general health (as measured by 36-Item Short Form Health Survey). Biological samples: Peripheral whole blood samples were collected at baseline, 3-month, and 6-month follow-up for RNA sequencing and DNA methylation. RNA-seq was conducted on two Illumina NovaSeq 6000 S4 flow cell lanes using a paired-end 2 × 150 bp configuration, and transcript quantification was performed with Salmon (reference genome build: GRCh38). Statistical analysis: All analyses were performed in R (version 4.5.1). Differentially expressed gene (DEG) analysis was performed for protein-coding genes using negative binomial generalized linear models as implemented in DESeq2, adjusted by age and sex. For each treatment, expression differences were examined compared to baseline at 3- and 6-month follow-up and between depression remitters and non-remitters (HAM-D ≤ 6). Pathway enrichment analysis was conducted using the fgsea algorithm using the Human Molecular Signatures Database Hallmark gene sets. Results with Benjamini-Hochberg adjusted p-value (FDR) < 0.05 were considered statistically significant for all analyses.

Results: At 3-months, the HEW treatment transcriptional profile was marked by significant (adj.p < 0.05) upregulation of the Interferon Alpha Response pathway only, with no significant pathway enrichments noted at 6-month follow-up. By contrast, at 3- and 6-month follow-ups, the TCC treatment transcriptional profile revealed significant downregulation of the Interferon Gamma Response and Interferon Alpha Response. When examining depression remitters vs. non-remitters for each treatment, significant downregulation of Interferon Gamma Response and Interferon Alpha Response signaling pathways were noted for TCC at 3-month follow-up but not for HEW. At 6-month follow-up, TCC and HEW showed significant opposing enrichment patterns for Inflammatory Response, P53, and Apoptosis signaling pathways—upregulated in HEW and downregulated in TCC.

Conclusions: TCC downregulated interferon signaling pathways at both 3- and 6-month follow-ups, an effect linked to depression remission, and further demonstrated sustained suppression of inflammatory response, p53, and apoptosis pathways—contrasting with upregulation of these same pathways in HEW. These results suggest that TCC may promote remission of late-life depression by modulating immune, stress, and cell survival signaling, highlighting its potential as a biologically active, integrative intervention for treatment-resistant geriatric depression.

Keywords: geriatric depression, treatment-resistant-depression, integrative neuroscience, transcriptome biology, bioinformatics

Disclosure: Nothing to disclose.

P200. FUNCTIONAL NEUROIMAGING CORRELATES SPECIFIC TO ANXIETY, ANHEDONIA, AND GENERAL DISTRESS

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Background: Depression is a heterogeneous disorder in which symptoms such as anhedonia, anxiety, and general distress often overlap but may arise from distinct neural mechanisms. Anhedonia has been linked to reduced striatal and prefrontal activation during reward anticipation and outcomes, consistent with dopaminergic dysfunction. Anxiety, in contrast, is associated with heightened amygdala, insula, and dorsal anterior cingulate responses to threat and uncertainty, while general distress reflects broader disruptions in stress- and emotion-related circuits. Although anhedonia and anxiety both involve frontolimbic dysfunction, these occur in different contexts (e.g. reward versus threat) suggesting overlapping but dissociable neural signatures. Prior work has often conflated these symptom dimensions and relied on cross-sectional designs, limiting the ability to distinguish enduring vulnerability markers from transient state effects. Yet evidence that blunted striatal activation persists beyond acute episodes highlights the need to identify stable neural abnormalities. Differentiating the neural bases of symptom domains that frequently co-occur is also critical for advancing precision treatment. Longitudinal modeling of symptoms offers a powerful approach, enabling latent constructs that capture stable features to be isolated from measurement error and momentary fluctuations. The present study integrates symptom data collected across five timepoints with baseline functional magnetic resonance imaging (fMRI) of reward and stress processing. We test whether corticostriatal and cortico-limbic activation are uniquely associated with latent dimensions of anhedonia, anxiety, and general distress. By isolating shared versus distinct neural signatures, this design aims to clarify mechanisms underlying depression heterogeneity and inform the development of more targeted, mechanism-based interventions.

Methods: Data were drawn from a longitudinal study of college students examining neural mechanisms of stress coping and resilience. The present study sample included 171 college students; power calculations indicated an ability to detect moderate correlations at 0.4 using an alpha significance level of 0.05. Participants completed MASQ surveys at 0, 3, 6, 9, and 12 months and completed a functional magnetic resonance imaging battery at baseline. The Dice Task elicited corticostriatal activation during reward and corticolimbic activation during stress. Imaging data were preprocessed with the HCP minimal pipeline. Contrasts including 1) Reward > Neutral and 2) Stress > Neutral during 1) reward anticipation, 2) reward outcome, 3) stress anticipation and 3) stress outcome were modeled. Latent constructs of anhedonia, anxiety, and general distress were derived across five timepoints using structural equation modelling (SEM). Group-level analyses regress activation in corticostriatal and corticolimbic regions of interest (ROI) on these latent dimensions to test whether unique patterns of neural response to reward and stress are associated with specific symptom domains after accounting for shared variance. This design allows for a refined parsing of overlapping but dissociable neural mechanisms in internalizing psychopathology.

Results: ROI analyses indicated that anxiety was linked to decreased corticolimbic activation during stress anticipation ($\beta = -0.36$, $SE = 0.16$, $p = 0.026$). Whole-brain analyses further showed that anxiety was associated with reduced dorsolateral prefrontal cortex (dlPFC) activation during stress anticipation (TFCE-corrected). Anhedonia was not significantly associated with

corticostriatal reward response in this sample, and there were no whole-brain effects that survived correction.

Conclusions: Anxiety symptoms were associated with reduced corticolimbic activation during stress anticipation and diminished prefrontal cortex recruitment, consistent with models of altered affective responsivity and weaker regulatory engagement. These associations were specific to anxiety after accounting for anhedonia and general distress, highlighting the importance of parsing symptom dimensions when examining neural correlates of depression. In contrast, anhedonia was not linked to corticostriatal reward responses, which may reflect the use of latent constructs across multiple assessments or characteristics of a non-clinical sample. Overall, these findings provide preliminary evidence for distinct neural patterns tied to anxiety, while emphasizing the need for future research in clinical populations and with repeated imaging to determine whether such associations represent enduring vulnerability markers or context-dependent effects.

Keywords: Adolescent Depression, Reward, motivation and Anhedonia, Anxiety and stress, Neuroimaging Analysis, Clinical Neuroscience

Disclosure: Nothing to disclose.

P201. ANTIDEPRESSANT EFFICACY OF BLIXEPRODIL (GM-1020), A NOVEL, ORALLY ADMINISTERED NMDA RECEPTOR ANTAGONIST, IN A DOUBLE-BLIND, RANDOMIZED, PLACEBO-CONTROLLED STUDY IN PATIENTS WITH MAJOR DEPRESSIVE DISORDER

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Background: Blixeprodil (GM-1020) is a first-in-class, orally bioavailable N-methyl-D-aspartate (NMDA) receptor antagonist that combines a rapid onset of action and favorable tolerability with a convenient route of administration. It is currently being investigated as a rapid-acting antidepressant for the treatment of major depressive disorder (MDD). In this study, we investigated the safety and efficacy of GM-1020 in patients with recurrent MDD.

Methods: This proof-of-concept study had two parts: Part A was a randomized, double-blind, placebo-controlled, single-dose, cross-over study of blixeprodil. In Part A Period 1, patients received blixeprodil 140 mg p.o. or placebo (PBO) and were monitored for 2 weeks. Participants then received the cross-over treatment and were monitored for another 2 weeks (Part A Period 2). Part B was a multiple-dose extension period; patients remained blinded to the treatment assignment and were randomized to receive blixeprodil at either 140 or 210 mg p.o., twice a week for 2 weeks. After Part B, all subjects were followed for an additional 28 days.

Subjects with recurrent MDD with Montgomery-Asberg Depression Rating Scale (MADRS) score ≥ 23 and a CGI ≥ 4 were randomized. Participants had either been antidepressant-free for at least 6 weeks or were receiving an adequate dose of a single SSRI or SNRI for at least 6 weeks, prior to screening and had to maintain this treatment unchanged during the study. SAFER interviews verified MDD diagnosis and severity and antidepressant medication status. The pre-specified exploratory efficacy endpoint was the change in MADRS total score from baseline to post-dose visits. Mixed model repeated measures (MMRM) methods were used to compare the different conditions (blixeprodil 140 mg vs PBO in Part A, 140 mg vs 210 mg in Part B). The analysis plan predefined that in the case of observed carryover effects in Part A Period 2, analyses of changes during Part A Period 1 only and/or

relevant subgroups (those with Part A Period 2 baseline MADRS total scores of ≥ 20) would be performed.

Sedation and dissociation were measured using the Modified Observer's Assessment of Alertness/Sedation scale (MOAA/S) and the Clinician Administered Dissociative State Scale (CADSS), respectively.

Results: Forty-six subjects (mean age of 35.4 years, 22 female, mean baseline MADRS of 32.3) were randomized and received at least one dose of study drug. As a carryover effect was observed in Part A Period 2, Part A Period 1 only analyses were performed. In Period 1, bupropion 140 mg exerted a strong antidepressant effect compared to placebo at 24 h post-dose (LS Mean (LSM) difference = -6.3 ; $p = 0.006$, effect size (ES) = 0.85) and at Day 8 (LSM difference = -5.1 ; $p = 0.076$, ES = 0.55). At the start of Part B, mean MADRS scores were approximately 10 points lower than those at baseline in Part A, and patients showed further improvement during this dosing extension period. No significant differences were observed between bupropion 140 and 210 mg at any Part B timepoint. After the final dose, the antidepressant response remained durable, with minimal attenuation during the 28-day follow up. Importantly, the remission rates for the combined arms were 53% at the end of Part B (Day 42), and 58% at the end of follow-up (Day 67). These findings suggest a robust and durable antidepressant effect of bupropion.

Both dose levels were found to be safe and well-tolerated with respect to TEAEs (no SAEs and no TEAEs leading to discontinuation of treatment), blood pressure increases, and with minimal effects on dissociation (CADSS) and sedation levels (MOAA/S).

Conclusions: These results suggest that the novel orally bioavailable NMDA receptor antagonist bupropion exerts clinically relevant, statistically significant, and rapid antidepressant effects after a single dose and durable antidepressant effect after biweekly administration. Bupropion was well tolerated and demonstrated a favorable safety profile with low dissociative potential in patients with MDD. Further studies investigating the efficacy, safety, and tolerability of bupropion are warranted and planned.

Keywords: Novel Rapid Antidepressant, Major Depressive Disorder (MDD), NMDA Antagonists, Randomized Double-Blind

Disclosure: Gilgamesh Pharmaceuticals, Employee, Self, Gilgamesh Pharmaceuticals, Stock/Equity - Privately Held Company, Self.

P202. LONGITUDINAL PATTERNS OF BRAIN AND CARDIOVASCULAR RESPONSES TO ACUTE STRESS IN REMITTED AND RECURRENT LATE-LIFE DEPRESSION

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Background: In remitted late-life depression (LLD), recurrence is common and is associated with adverse health outcomes, including premature mortality. A key risk factor for LLD and its recurrence may involve dysregulated reactivity to acute psychological stressors. We previously reported cross-sectionally that recently remitted LLD show blunted brain and cardiovascular reactivity to stress. What is unclear, however, is whether these initial cross-sectional differences pertaining to remitted and recurrent LLD persist over longitudinal follow-up of up to 2 years, and whether they reflect stable vulnerability markers or otherwise reflect recent fluctuations in depression severity.

Methods: Participants were recruited through 3 sites and included 145 remitted LLD (60 to 82 years; 70% female; 87% white) and 63 age-matched healthy control (HC) adults (60 to 79 years; 62% female; 78% white). Participants underwent a validated

acute stressor task – the modified Multi-Source Interference Task (MSIT) – during fMRI with cardiovascular monitoring at baseline testing and upon repeat testing at 8, 16, and 24 months. LLD participants were clinically interviewed at each testing visit to evaluate depression severity and remission status. Linear mixed-effects models tested for effects of group, visit, and their interaction on behavioral, affective, cardiovascular, and brain responses to the MSIT, adjusting for age, sex, site, and cardiovascular risk.

Results: Participants in the analytic cohort completed $M \pm SD = 2.92 \pm 1.12$ study visits (608 total). In the LLD group, 77 (53%) relapsed (REL) at some point during the longitudinal follow-up period, as opposed to maintaining stable remission (REM). There were stable group differences in several other outcomes: compared to HC, LLD exhibited slower reaction times ($t = -3.36$, $p = 0.019$), more pronounced affective valence shifts ($t = 3.27$, $p = 0.002$), reduced stressor-evoked systolic blood pressure (SBP; $t = -2.93$, $p = 0.004$) reactivity, and reduced stressor-evoked BOLD signal activity in the amygdala ($t = -2.92$, $p = 0.004$), dorsolateral prefrontal cortex (DLPFC; $t = -2.34$, $p = 0.02$), dorsal anterior cingulate (dACC; $t = -2.45$, $p = 0.015$), and posterior insula ($t = -2.83$, $p = 0.005$). None of these outcomes showed significant interactions with visit. There was a group-by-visit interaction on heart rate (HR) reactivity ($F = 3.46$, $p = 0.017$), wherein group differences were only significant at the baseline visit ($t = -3.06$, $p = 0.003$), and not at any follow-up visits (p 's > 0.64). When comparing HC to relapsers (REL) and stable remitters (REM) among these outcomes, several similar findings emerged, with stable significant group differences specific to the REL group in terms of reduced stressor-evoked SBP ($t = -2.85$, $p = 0.013$), DLPFC ($t = -2.64$, $p = 0.0024$), dACC ($t = -2.78$, $p = 0.02$), and posterior insula reactivity ($t = -2.86$, $p = 0.01$).

Conclusions: These longitudinal findings add important context to our prior cross-sectional report and suggest that several features of stress reactivity in LLD may reflect stable characteristics persisting beyond acute episodes and are more pronounced in participants who eventually relapse. Initial group differences in HR reactivity may normalize upon repeat testing, perhaps due to well-known patterns of habituation often observed in healthy participants yet not observed in this LLD cohort. By contrast, Hence, they may represent a neurophysiological vulnerability marker for depression relapse in late life. Future work will examine to what extent patterns of stress reactivity more proximally predict subsequent relapse or sustained remission in LLD.

Keywords: Acute Stress, BOLD fMRI signal, Late Life Depression, Sustained remission, Brain and cardiovascular responses

Disclosure: Nothing to disclose.

P203. EVIDENCE FOR INDIVIDUALIZED DIGITAL ENGAGEMENT WITH SMARTWATCH DATA TO REDUCE DEPRESSIVE SYMPTOM SEVERITY

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Background: Although smartwatches offer the potential for real-time stress monitoring and adjunctive support in depression management, little research has assessed their role in improving depression severity in individuals, and long-term (12-month) longitudinal studies are notably absent.

Methods: Participants in a 12-month decentralized wellbeing study at the Mayo Clinic wore a commodity smartwatch as a digital health technology (DHT) and completed electronic surveys on mental and overall functioning at baseline and on a quarterly basis. The Center for Epidemiological Studies – Depression (CES-D) scale was used to measure depression symptom severity. Participants were requested to wear the watch $\geq 70\%$ of the time and received individualized quarterly summaries of their smartwatch data that included average walking distance (monthly and quarterly), daily sleep durations, heart rate (minimum, maximum, and median), and a subset of participants had access to their heart rate variability (HRV). Individuals with stable depression were those with baseline CES-D total score < 16 and reported current antidepressant use. Non-depressed controls had a CES-D total score < 16 and no reported antidepressant use. A 4 point or greater decrease in the CES-D total score from baseline to 12-months was considered a clinically meaningful reduction in depressive symptoms. Linear mixed-effects models were used to examine differences in functioning (with watch adherence $> 50\%$) between those with stable depression and non-depressed controls, with participants included as random effects and sex, age, race, and marital status as covariates. Logistic regression was using to examine the differences in clinically meaningful change between stable depressed participants and non-depressed controls.

Results: Of 531 participants, 436 (82%) completed the study and 362 (68%) completers wore the watch $\geq 70\%$ of the time, resulting in a total of 194 million minutes of smartwatch data. Among completers, 48 had stable depression and 55 were non-depressed controls ($n = 103$ total). Compared to those with stable depression, non-depressed controls walked 1.23 miles more each day ($p < 0.001$), slept 25 minutes less each night ($p < 0.006$), had 22 min less light sleep ($p < 0.002$), and 5 minutes less awake time ($p < 0.04$). Seventy-nine subjects had HRV data, and HRV in non-depressed controls improved by 9ms ($p < 0.009$) in comparison with those with stable depression. The odds of a clinically meaningful reduction in CES-D total score among non-depressed controls was 3.63 ($p = 0.05$, CI: 1.06–13.99) in comparison with those with stable depression.

Conclusions: Individualized digital data engagement when paired with easily accessible DHT such as smartwatches may provide a scalable means to promote behavior activation and track digital biomarkers relevant to general health and mood state over the long term.

Keywords: Depression, Digital Biomarkers, wearable technology, physical activity, sleep

Disclosure: Nothing to disclose.

P204. NEW ANTIDEPRESSANT USE AMONG COMMUNITY HEALTH CENTER PATIENTS WITH DEPRESSION DURING AND AFTER THE COVID-19 PANDEMIC

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Background: From 2021 to 2024, community health centers (CHCs) served 30 to 34 million patients at more than 17,000 locations, an increase of 12% over 4 years. During this time, over 30% of all people living in poverty and 20% of US rural residents were CHC patients. Since the COVID-19 pandemic, CHC patients overall report an increased mental health visits and negative mental health outcomes. However, from 2019 to 2021, paradoxically, we and others reported decreased rates of new antidepressant prescriptions among CHC patients. We report the continued study of these patient cohorts to gain further insights into mental health care access and practice to drive the development of future interventions.

Methods: We conducted an observational, retrospective, exploratory study using data from a CHC electronic health record (EHR). Patients were at least 18 years old, had a diagnosis of depression, and were prescribed a new antidepressant During (March 2020 to February 2021) and After (March 2023 to February 2024) the COVID-19 pandemic. We assessed patient profiles for demographics, mental health diagnosis, new and follow up antidepressant medication prescriptions, general and preventative risk assessments, depression screenings, and comorbid conditions. From 2020 to 2021 and 2023 to 2024, the CHC annual census of adult patients was 5927 and 6541, respectively. The primary study endpoint was the number and proportion of adult CHC patients with a new antidepressant prescribed for each cohort. Secondary endpoints were use of mental health services, preventative (social determinants of health) and depression screenings, antidepressant medication components, and antidepressant medication changes (reorder, dose increase, and discontinuation reason). Descriptive and inferential statistics were used to tally and compare within and between groups. This study was exempt from University of Michigan Institutional Review Board review (HUM#00195619).

Results: For the During COVID-19 cohort, 500 adult patients with depression started a new antidepressant medication. Of these, 72% were female and 36% were White; the average age was 49 ± 14 years old and 57% preferred to speak Spanish. For the After COVID-19 cohort, 638 patients met study criteria; of these, 70% were female, and 30% were White; the average age was 48 ± 14 years old and 61% preferred to speak Spanish. From 2020 to 2024, the number of adults with a new antidepressant prescription increased by 21.6%. During this time period, the CHC's adult patient census increased by 9.4%.

For both cohorts, the most common associated diagnoses (composites) were depression or anxiety. Over half of patients in both cohorts had associated diagnoses of pain, insomnia, and other chronic conditions. Sertraline was the most commonly prescribed antidepressant (28% and 24%, During and After cohorts, respectively) at the 25 mg starting dose. Most patients were prescribed a 30-day supply with an average of 3 refills, followed by a 90-day supply and 0–1 refills. The majority (60–65%) of the antidepressants were reordered. Additionally, most were initially prescribed and then continued at the respective antidepressant recommended starting dose. The most common reasons reported for antidepressant discontinuation were "inactive medication," "patient choice," and "ineffective medication." Few patients were prescribed a subsequent alternative antidepressant or adjunct therapy, other than behavioral health services. Overall, the CHC reported an increase in mental health screening and follow up; however, these were not proportionately observed in the new antidepressant cohorts. Furthermore, without timely depression screening, depression symptom improvement and subsequent remission rates from depression could not be assessed. Study limitations include retrospective design, and EHR as data source.

Conclusions: This ongoing exploratory study of new antidepressant prescriptions among CHC adult patients identified key gaps in depression screening, antidepressant titration, adjunct therapy, and follow up within and between the During and After COVID-19 cohorts. Future analyses are needed and should include COVID-19 related comorbid conditions that may provide insight into the differences noted between cohorts and expected new antidepressant prescription and follow up.

Keywords: Depression, Health services, Mental health outcomes, Antidepressant, Community health center

Disclosure: Nothing to disclose.

P205. SEX-SPECIFIC MITOCHONDRIAL ALTERATIONS IN COGNITIVELY UNIMPAIRED OLDER DEPRESSED INDIVIDUALS

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Background: Late-life major depressive disorder (MDD) is a leading cause of disability and contributes to accelerated cognitive decline in older adults. It is also considered a risk factor for the development of Alzheimer's disease (AD).

Biological aging processes, including mitochondrial dysfunction, are increasingly recognized as key contributors to the pathophysiology of late-life depression and AD. Mitochondria regulate energy production, stress responses, and apoptosis in neurons and glia, while dysfunctional mitochondria can trigger pro-inflammatory signaling and cellular senescence. Circulating mitochondrial DNA (mtDNA), present as cell-free fragments (ccf-mtDNA) or encapsulated within extracellular vesicles (EV-mtDNA), has emerged as a non-invasive biomarker of mitochondrial stress. However, the interrelationships among peripheral mitochondrial biomarkers and depression status in late-life depression remain poorly understood.

This led us to examine whether depression or depression severity influences plasma levels of ccf-mtDNA and EV-mtDNA. In addition, given that females are more likely to be diagnosed with depression and AD, we investigated the influence of sex on these associations.

Methods: This study is a secondary analysis of an observational cohort of cognitively unimpaired older adults with and without MDD ($N = 112$) who were followed for three years. At baseline, all participants underwent a clinical evaluation that included a physical and neurological examination, standardized clinical scales, cognitive testing, and a blood draw for plasma collection. The diagnosis of major depressive disorder was established through psychiatric evaluation in conjunction with the Structured Clinical Interview for DSM-IV.

ccf-mtDNA and EV-mtDNA were determined using quantitative polymerase chain reaction (qPCR).

We examined the relationship between depression status (MDD vs. controls) and mitochondrial function, as well as plasma levels of ccf-mtDNA and EV-mtDNA, using independent-samples t-tests. We also tested for interactions between MDD status and sex on mitochondrial function using two-way analysis of variance. Finally, Pearson's correlations were conducted to assess associations between mtDNA variables and depression status.

Results: Group-level analyses showed elevated levels of both EV-mtDNA and ccf-mtDNA in the depressed group compared with controls. Across the entire sample, depression severity was positively associated with both measures. Across all participants, we observed moderately strong correlations ($p < 0.001$) between plasma levels of ccf-mtDNA and EV-mtDNA. However, these associations appeared to be driven by the control group, as none were significant within the MDD group alone. We examined interactions between key measures of ccf-mtDNA and EV-mtDNA. No significant interactions were found between diagnosis and genotype or between sex and genotype. However, significant interactions emerged between diagnosis (depressed vs. controls) and sex (female vs. male) for both ccf-mtDNA and EV-mtDNA. Older depressed females exhibited higher EV-mtDNA levels than older depressed males, whereas older depressed males showed higher ccf-mtDNA levels than their female counterparts.

Conclusions: These findings suggest that mitochondrial alterations in depression may be shaped by sex-specific biological pathways in the aging population. Moreover, targeting

mitochondrial function could represent a promising approach to managing depression in older adults.

Keywords: Depression, Circulating cell-free mitochondrial DNA, mitochondrial dysfunction

Disclosure: Nothing to disclose.

P206. BEYOND PASSIVITY: DEPRESSIVE SYMPTOMS PREDICT PERSISTENT ACTIVE AVOIDANCE UNDER AMBIGUITY

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Background: Avoidance behaviors can involve either initiating or suppressing actions to prevent harm. While avoidance generalization has been extensively studied in anxiety, its role in depression remains less clear. In particular, it is unknown how depressive symptoms bias action selection when active and inhibitory avoidance strategies directly compete under ambiguity. To test competing accounts of depression—one emphasizing behavioral passivity, the other behavioral rigidity—we developed a novel behavioral task to assess avoidance generalization and see how this may relate to depressive symptoms.

Methods: The avoidance generalization task was conducted online with minimal instructions to promote instrumental learning and parallel rodent paradigms of avoidance (Dalton et al., 2025). Discriminative cues signaled either active (button presses) or inhibitory (withholding) responses to prevent aversive sounds, introduced across three stages: acquisition (active only), intermixed (active/inhibitory), and bias (active/inhibitory with probe trials). In the bias stage, probe trials presented innocuous generalization stimuli (GSs) varying in similarity to discriminative cues, progressing from active-like to inhibitory-like across blocks. This design allowed us to test whether depressive symptoms favored behavioral passivity or behavioral rigidity under ambiguity. Using a dimensional approach we recruited undergraduates ($N = 292$; both sexes included) presenting with a wide range of depressive symptom scores (BDI-II; minimal-severe). Linear mixed models (LMMs) assessed relationships between depressive symptoms and avoidance, including active and inhibitory accuracy as well as probe trial responding.

Results: Participants readily acquired both avoidance strategies. Elevated depressive symptoms predicted (1) greater maintenance of active avoidance once responses were established ($\beta's > 0.112$, $p's < 0.04$), and (2) a stronger bias toward active responding to ambiguous cues ($\beta = 0.376$, $p = 0.03$), even when these cues more closely resembled inhibitory signals. No significant depressive symptom effects were observed during initial acquisition ($\beta = -0.026$, $p = 0.63$).

Conclusions: These findings challenge traditional views of depression as primarily reflecting motivational deficits or behavioral passivity. Instead, in the specific context of our task, they highlight depressive symptoms can promote persistent, effortful avoidance under ambiguity, consistent with behavioral inflexibility and reduced cognitive control. By directly contrasting active and inhibitory avoidance in a generalization context, our paradigm provides a mechanistic tool for refining models of aversively motivated behavior in psychiatric disorders.

Keywords: persistent avoidance, Active Avoidance, Ambiguity, dimensional, depressive symptoms

Disclosure: Nothing to disclose.

P207. ASSOCIATIONS BETWEEN SUBJECTIVE AND OBJECTIVE FATIGUE AND COGNITIVE FUNCTIONING IN INDIVIDUALS WITH ANXIETY AND MOOD DISORDERS

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Background: Fatigue is a common complaint among individuals with anxiety and mood disorders (AMD) [1]. Subjective fatigue is closely related to cognitive functioning in healthy adult population [2], as well as in other populations, such as individuals with coronary artery disease [3]. Even though cognitive dysfunction and fatigue are both more common in individuals with AMD than in otherwise healthy individuals [4], the relationship between fatigue and cognitive functions is still underexplored. Furthermore, most research on fatigue relies primarily on self-report measures, often lacking objective assessments to compare with. Therefore, the aim of our current study was to investigate the correlations between subjective and objective fatigue and cognitive functions in individuals with AMD.

Methods: Three hundred and fourteen individuals (77.7% women and 22.3% men, mean age 40.2 ± 11.9 years) attending a Stress-related disorders day care unit of the Palanga Hospital at Neuroscience Institute of the Lithuanian University of Health Sciences, participated in this cross-sectional study. Diagnosis of AMD was established using the Mini-International Neuropsychiatric Interview. Subjective fatigue was assessed using the Multidimensional Fatigue Inventory-20 (MFI-20), while objective fatigue was evaluated via an exercise capacity (EC) workload test. Cognitive functioning was measured using Digit Span Test, Digit Symbol Test, Trail Making Test (TMT) Parts A and B. Depression and anxiety symptoms were evaluated with The Patient Health Questionnaire-9 (PHQ-9) and Generalized Anxiety Disorder-7 (GAD-7) scale. Associations between fatigue and cognitive functioning were first analysed using univariate regression. Multivariable regression analyses were then conducted, adjusting for age, sex, education, current medication use, history of smoking, depression and anxiety symptoms.

Results: In individuals with AMD, univariate regression analyses found associations between subjective and objective fatigue characteristics and cognitive functions. Specifically, higher scores on the MFI-20 subscales for reduced activity and reduced motivation were associated with lower T-scores on TMT Part A ($\beta = -0.117$, $p = 0.039$; $\beta = -0.112$, $p = 0.048$ respectively); higher EC was associated with higher numbers of pairs recalled correctly in Digit Symbol Test ($\beta = 0.195$, $p = 0.001$), higher TMT Part A T-score ($\beta = 0.156$, $p = 0.011$), and higher TMT Part B T-score ($\beta = 0.125$, $p = 0.041$). After completing multivariable regression models adjusting for age, sex, education, current medication use, history of smoking, depression and anxiety scores, only the association between higher EC and higher number of pairs recalled correctly in Digit Symbol Test remained statistically significant ($\beta = 0.227$, $p = 0.002$). Additionally, multivariable regression analysis found that higher EC was independently associated with higher TMT A/B T-scores ($\beta = 0.152$, $p = 0.046$).

Conclusions: Overall, in individuals with AMD, objective fatigue, as measured by EC, was independently associated with poorer cognitive functioning, particularly in domains involving incidental learning, recall, and executive functioning. These findings underscore the importance of distinguishing between subjective and objective fatigue measures and their respective impact on cognitive performance. These findings have practical implications for better understanding the relationship between cognitive functions and fatigue in individuals with AMD and provide the

basis for future research investigating the relationship between cognitive function and fatigue.

Keywords: Anxiety and Depression, mood and anxiety disorders, fatigue, Cognitive Functioning

Disclosure: Nothing to disclose.

P208. DISCOVERY OF A NOVEL GENE-DRUG INTERACTION BETWEEN DESVENLAFAXINE AND UGT2B15 IN A COHORT OF LATE-LIFE DEPRESSED PATIENTS TREATED WITH VENLAFAXINE

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Background: Venlafaxine, a common SNRI (Serotonin Norepinephrine Reuptake Inhibitor), is metabolized by phase I CYP450 enzymes into its main metabolite desvenlafaxine. Desvenlafaxine, also available as an independent therapeutic, is indicated for major depressive disorder (MDD) and, while less commonly prescribed than other antidepressants, may be an important therapeutic option for patients who have failed previous antidepressant treatment. Given the wide therapeutic range and variability of clinical response to desvenlafaxine, pharmacogenetic factors may inform desvenlafaxine treatment. Desvenlafaxine is metabolized via phase II metabolism, specifically glucuronidation, which is mediated by the UDP-glucuronosyltransferase (UGT) superfamily of enzymes. Glucuronidation is the major route of elimination of desvenlafaxine, therefore, genetic variation in this pathway may influence drug efficacy and toxicity. The specific isoform(s) within the UGT family responsible for desvenlafaxine glucuronidation have not yet been identified. The aim of this study was to identify and characterize potential pharmacogenetic interactions between 10 variants across seven different UGT isoforms and desvenlafaxine plasma concentrations.

Methods: In the Incomplete Response in Late-Life Depression: Getting to Remission (IRL-Grey; NCT00892047) study, DNA samples and plasma concentrations from older adults (age ≥ 60) with depression who were being treated with venlafaxine XR were analyzed. Plasma concentrations of venlafaxine and desvenlafaxine were measured via tandem mass spectrometry at two timepoints, after 4 and 12 weeks of treatment. In this study, patients were genotyped for UGT1A1 (rs35350960), UGT1A3 (rs3821242 and rs45449995), UGT1A4 (rs2011425), UGT1A6 (rs1105880 and rs2070959), UGT1A9 (rs145084767 and rs72551330), UGT2B7 (rs7439366), and UGT2B15 (rs1902023, commonly referred to as *2). In the statistical analysis, dose-corrected, log10-transformed 1) desvenlafaxine and 2) total active moiety (sum of venlafaxine and desvenlafaxine) plasma concentrations were regressed on each UGT genotype using a mixed-effects model for repeated measures. Covariates adjusting for age, sex, race, ethnicity, BMI, visit, and time since last dose were also included in the models. CYP2D6 metabolizer status was also used as a covariate because CYP2D6 is the major metabolizing enzyme responsible for forming desvenlafaxine from venlafaxine. Percent changes were calculated from estimated marginal means. P-values were considered significant at $p < 0.05$ and were not corrected for multiple testing, due to the hypothesis-generating nature of this study.

Results: 278 patients had complete data and were included in analyses. Of the 10 variants evaluated, only UGT2B15*2 (rs1092023) was significantly associated with desvenlafaxine plasma concentrations ($F = 7.0$, $p = 0.001$). Compared to patients without UGT2B15*2 ($n = 71$), homozygous *2 patients ($n = 88$) had 26% higher desvenlafaxine plasma concentrations and

heterozygous *2 patients ($n = 119$) had 28% higher desvenlafaxine plasma concentrations. UGT2B15*2 was also significantly associated with dose-corrected active moiety plasma concentrations ($F = 5.6$, $p = 0.004$). Compared to patients without *2, active moiety plasma concentrations were 19% higher in homozygous *2 patients and 21% higher in heterozygous *2 patients.

Conclusions: This was the first comprehensive study that analyzed variants across many UGT isoforms and observed a meaningful gene-drug interaction with desvenlafaxine plasma concentrations. UGT2B15*2 was associated with higher desvenlafaxine plasma concentrations in a cohort of older adults with depression treated with venlafaxine. This study suggests that UGT2B15 may be involved in desvenlafaxine metabolism and presents a novel discovery of a gene-drug interaction for desvenlafaxine. Future directions could include replication of the finding in an independent cohort, as well as investigation of the clinical impact of reduced clearance of desvenlafaxine in UGT2B15*2 allele carriers.

Keywords: Antidepressants, Major depressive disorder (MDD), Pharmacokinetics, Genetic biomarker

Disclosure: Myriad Genetics, Employee, Self, Myriad Genetics, Stock / Equity - Publicly Traded Company, Self.

P209. Poster Withdrawn

P210. REDUCTION IN C-REACTIVE PROTEIN FOLLOWING INFILIXIMAB IS ASSOCIATED WITH INCREASED REWARD AND PROBABILITY SENSITIVITY IN DEPRESSION

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Background: Growing evidence suggests that inflammation may contribute to the pathophysiology of motivational anhedonia in major depressive disorder (MDD). However, few studies have directly examined how elevated inflammation impacts motivated behavior in depression. Here, we present novel findings from a randomized clinical trial (NCT03006393) investigating the mechanisms by which inflammation contributes to motivational anhedonia in MDD, and how treatment with the TNF- α antagonist infliximab affects motivated behavior. Prior work from this trial demonstrated enhancement in prospective effort-based decision-making using an fMRI-based task in which individuals made choices and completed the effort separately (Treadway et al., 2025). In this study, we examined the effects of infliximab on a second effort-based decision-making task for which individuals were required to expend effort on each trial.

Methods: Thirty patients (age 24–58, 87% female) with C-reactive protein (CRP) levels >3.0 mg/L received a single infusion of either infliximab (5 mg/kg) or saline in a randomized, double-blind, placebo-controlled trial. Peripheral inflammatory markers were assessed at baseline and 14 days post-infusion. Motivated behavior at baseline and 7 days post-infusion was evaluated using the Effort Expenditure for Rewards Task (EEfRT), a multi-trial decision-making task in which participants made repeated choices between a low-effort option and a high-effort option of varying reward value and probability of receipt (low, medium, high). Generalized linear mixed models were used to examine baseline associations between CRP, reward, and probability level on choice in the EEfRT, and the effects of treatment and CRP change on these measures.

Results: At baseline, significant CRP $\times$ Probability effects on choice were observed, such that higher CRP levels were associated with a reduced effect of probability on choice at both medium

($B = -0.644$, $z = -3.204$, $p = 0.001$) and high levels of probability ($B = -0.578$, $z = -2.889$, $p = 0.004$). Following treatment, significant CRP Change $\times$ Treatment Arm $\times$ Probability $\times$ Time ($B = -1.659$, $z = -2.289$, $p = 0.022$ [medium]; $B = -1.659$, $z = -2.605$, $p = 0.009$ [high]) and CRP Change $\times$ Treatment Arm $\times$ Reward $\times$ Time interactions ($B = -0.754$, $z = -2.599$, $p = 0.009$) were observed, indicating that patients with the greatest reduction in CRP exhibited increased sensitivity to reward and probability following infliximab treatment.

Conclusions: Consistent with our prior work showing that infliximab increased willingness to expend effort relative to placebo, the current study found that infliximab treatment enhanced reward- and probability-based effort allocation as a function of reduced inflammation. These findings suggest that improvements in motivated behavior are linked to decreases in peripheral inflammation, supporting a mechanistic role for inflammation in motivational anhedonia across multiple effort-based tasks.

Keywords: Depression, inflammation, motivation, anhedonia

Disclosure: Nothing to disclose.

P211. BEYOND SELF-REPORT: DETECTING DEPRESSIVE SYMPTOMS DIRECTLY FROM NEURAL ACTIVITY

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Background: Depression is typically assessed through clinical interviews and self-report rating scales. These approaches, however, are vulnerable to self-presentation bias, cognitive load, and limited access to implicit negative schemas. Subtle or under-reported symptoms may therefore go undetected, particularly in groups facing stigma or cognitive impairment. There is therefore a pressing need for direct, sensitive, bias-resistant measures of symptom expression. We asked whether depressive symptoms could be detected directly from neural activity during natural reading, without requiring any overt judgments or introspection.

Methods: Forty adults spanning a wide range of depressive symptoms (indexed by the Beck Depression Inventory-II) read 160 self-relevant depression probes, specifically written to capture core constructs such as low mood, anhedonia, fatigue, guilt, poor concentration, hopelessness, and established risk factors including trauma and social isolation. Each vignette established a context reflecting core clinically relevant experiences and concluded with a first-person statement that was either depression-consistent (e.g., 'Lately my mind has been quite foggy') or depression-inconsistent (e.g., 'Lately my mind has been quite clear'). A matched set of third-person vignettes controlled for general sensitivity to depression-related content and expectancy effects. Neural responses to the critical words were recorded with EEG as participants read the probes naturally, without making explicit judgments.

Results: Depression severity predicted a sustained, highly selective neural response distinguishing self-relevant depression-consistent from depression-inconsistent statements. Between 300 and 500 ms, reduced neural activity to depression-consistent statements indicated that negative self-schemas influenced expectations at the earliest stage of comprehension. Between 500 and 1000 ms, larger neural responses to depression-consistent statements reflected greater attentional engagement and motivational salience. These neural signatures scaled continuously with symptom severity, even at subclinical levels, highlighting their

sensitivity to individual variation. Importantly, they were absent in non-self-relevant probes, demonstrating selectivity to personally relevant material and ruling out alternative explanations such as general emotional reactivity or differences in contextual prediction.

Conclusions: Depressive symptoms can be detected directly from neural activity during natural reading of self-relevant clinical probes, even in individuals without clinical diagnoses and without overt behavioral responses. This study provides the first demonstration that neural activity alone can reveal how DSM-defined depressive symptoms and risk factors are instantiated in real time during natural comprehension. By capturing how negative self-schemas are recruited online, this approach offers a mechanistic bridge between clinical constructs and brain function. With further development, such probes could form the basis of “neural rating scales” that complement traditional assessment tools, capturing what patients cannot or will not report. Such measures could provide bias-resistant markers for early detection, continuous symptom monitoring, and individualized treatment planning, advancing the broader goal of precision psychiatry.

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Keywords: precision psychiatry, EEG/ERP electrophysiology, depression schema

Disclosure: Nothing to disclose.

P212. BRAIN NETWORK-DRIVEN PREDICTIVE NEURAL MARKERS FOR SUICIDE RISK

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Background: Suicide is a major public health issue, yet its neurobiological underpinnings remain poorly defined. Multiple, distinct pathways can culminate in suicidal behavior, making it unlikely that any single variable will serve as a universal predictor. Consequently, the field needs objectively measurable biomarkers that capture the complexity of suicide risk. Resting-state functional connectivity within the default mode (DMN), executive control (ECN), and salience (SN) networks has emerged as a promising indicator of mood dysregulation and suicidal ideation. Most previous work has relied on hemodynamic measures, which capture neural activity on the scale of seconds and thus blur fast neural dynamics. Magnetoencephalography (MEG) resolves activity at the millisecond level and therefore preserves this temporal detail. When combined with parcel-based orthogonalized envelope correlations, resting-state MEG can quantify resting-state coherence (i.e., amplitude coupling between brain regions within a network) offering a direct, high-resolution probe of network integrity. We hypothesized that individuals in an acute suicidal crisis (suicide attempt or suicidal ideation within the past two weeks) would show reduced MEG-derived connectivity within the DMN, ECN, and SN compared with three comparison groups: (1) individuals with a lifetime history of suicide attempts but no current crisis, (2) depressed clinical controls without suicide risk, and (3) healthy controls without personal or familial psychiatric history. By integrating these neural metrics into a multi-omics

framework, the study aims to advance objective assessment of imminent suicide risk.

Methods: 77 participants (47F; 30M) were included in the study (mean age = 38.41 ± 14.09 years). The participant groups included: 1) those experiencing a recent suicidal crisis (high risk; n = 8); 2) those with a history of suicide attempts but not in the past year (lower risk; n = 26); 3) clinical controls with depression but no history of suicide risk (clinical controls; n = 22); and 4) individuals with no personal or family history of either psychiatric disorder or suicidal behavior (minimal risk; n = 21). Participants completed one or two eyes-closed resting-state recordings on a 275-channel CTF system. A T1-weighted MRI was acquired and co-registered via fiducial marks. MEG data were band-pass filtered (2–58 Hz), source-reconstructed, parcellated into 400 cortical regions, orthogonalized to reduce signal leakage, and Hilbert-transformed to extract amplitude envelopes. Data Integration Analysis for Biomarker discovery using Latent variable approaches for Omics studies (DIABLO) identified the connectivity features that best separated the high-risk group from all others. These features were submitted to ANCOVA (age, sex covariates) to test group differences; significant effects were probed with pairwise post-hoc comparisons. Because of non-normal distributions and small sample sizes, we then applied Bayesian generalized linear mixed models with Markov-chain Monte Carlo sampling to assess the association between current suicidal-ideation severity and the selected connectivity features.

Results: The DIABLO model reliably separated the four groups with 999 permutations ($p = 0.018$). Receiver-operating-characteristic curves confirmed good discrimination of the high-risk group from all others in each network (DMN AUC = 0.77; ECN AUC = 0.82; SN AUC = 0.80). The high-risk group exhibited significantly weaker functional connectivity within specific regions of these networks ($ps < 0.05$). Further analyses revealed that recent suicidal ideation scores were positively associated with functional DMN connectivity (e.g., between the precuneus/posterior cingulate cortex and dorsal/medial prefrontal cortex) and SN connectivity (e.g., between the medial frontal cortex and medial parietal cortex and between the parietal operculum and lateral prefrontal cortex) ($ps < 0.05$). Conversely, functional ECN connectivity (between the medial/posterior prefrontal cortex and precuneus) was negatively associated with recent suicidal ideation ($ps < 0.05$).

Conclusions: Our findings demonstrate that a multi-omics integration approach (DIABLO) can successfully extract electrophysiological biomarkers of suicide risk from resting-state MEG data. Individuals in an acute suicidal crisis showed markedly reduced connectivity within the DMN, ECN, and SN, and several of these network metrics were related to the severity of current suicidal ideation. These results provide preliminary evidence that MEG-derived network coherence may index imminent suicide risk. This study contributes to the growing understanding of neural mechanisms underlying suicidal thoughts, emphasizing the need for larger longitudinal studies with innovative research tools to validate these findings and improve clinical outcomes in this vulnerable population.

Keywords: magnetoencephalography, Suicide prediction, mood disorders

Disclosure: Nothing to disclose.

P213. IMPACT OF COMBINED CYP2D6 AND CYP2C19 METABOLIZER STATUS ON VENLAFAXINE METABOLISM IN OLDER ADULTS WITH DEPRESSION

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Background: Venlafaxine metabolism is strongly influenced by genetic variation in CYP2D6. Compared to CYP2D6 normal metabolizers (NM), poor metabolizers (PM) show elevated venlafaxine and lower metabolite (desvenlafaxine) plasma concentrations, while ultrarapid metabolizers are more likely to have lower venlafaxine and higher desvenlafaxine concentrations. While both venlafaxine and desvenlafaxine are equipotent, increased venlafaxine concentrations have been associated with an increased risk of side effects in CYP2D6 PMs. In addition to CYP2D6, several polymorphic enzymes may also contribute to plasma concentrations of both venlafaxine and its active moiety (sum of venlafaxine and desvenlafaxine). In this study, we asked whether genetic variation in CYP2C19, CYP2C9, and/or CYP3A4, in combination with CYP2D6 variation, could predict plasma concentrations of venlafaxine and its active moiety in a cohort of older adults with depression.

Methods: DNA and plasma concentrations from older adults (age ≥ 60) with depression and treated with venlafaxine XR in the Incomplete Response in Late-Life Depression: Getting to Remission study (IRL-Grey; NCT00892047) were analyzed. Venlafaxine and desvenlafaxine plasma concentrations were measured via tandem mass spectrometry at two timepoints, after 4 and 12 weeks of treatment. For this study, patients were genotyped for variants within CYP450 genes including CYP2C19, CYP2C9, CYP2D6, and CYP3A4, then categorized by metabolizer status for each gene. In the statistical analysis, dose-corrected, log₁₀-transformed 1) venlafaxine and 2) total active moiety plasma concentrations were regressed on CYP450 metabolizer status using a mixed-effects model for repeated measures. Covariates adjusting for CYP2D6 metabolizer status, age, sex, race, ethnicity, BMI, visit, and time since last dose were also included in the models. Percent changes were calculated from estimated marginal means.

Results: 278 participants had complete data and were included in analyses. In a multivariate model where CYP2D6 metabolizer status was the only CYP450 metabolizer status included, CYP2D6 was significantly associated with venlafaxine concentrations ($F = 37.3$, $p < 2.2 \times 10^{-16}$). When CYP2C19 metabolizer status was also included, both CYP2D6 and CYP2C19 were significantly associated with venlafaxine plasma concentrations ($F = 42.9$, $p < 2.2 \times 10^{-16}$ and $F = 5.7$, $p = 0.0008$, respectively). As CYP2D6 function decreased, venlafaxine plasma concentrations increased and, within each CYP2D6 metabolizer status, as CYP2C19 function decreased, venlafaxine plasma concentrations increased further. For example, compared to patients who were NM for both CYP2D6 and CYP2C19 (CYP2D6 NM/CYP2C19 NM, $n = 101$), CYP2D6 intermediate metabolizers (IM)/CYP2C19 NMs ($n = 76$) had 58% higher venlafaxine plasma concentrations while CYP2D6 IM/CYP2C19 IMs ($n = 19$) had 112% higher venlafaxine plasma concentrations. The largest increase in venlafaxine exposure was seen in a CYP2D6 PM/CYP2C19 PM ($n = 1$) who had an 879% higher venlafaxine plasma concentration compared to CYP2D6 NM/CYP2C19 NMs. Moreover, CYP2D6 PM/CYP2C19 NMs ($n = 20$) had 266% higher venlafaxine plasma concentrations compared to CYP2D6 NM/CYP2C19 NMs.

Active moiety was also analyzed in multivariate models. When CYP2D6 metabolizer status was the only CYP450 metabolizer status included in the model, it was not significantly associated with active moiety plasma concentrations ($F = 2.0$, $p = 0.11$). However, when metabolizer status for both CYP2D6 and CYP2C19 were included, both CYP2D6 and CYP2C19 were significantly associated with active moiety plasma concentrations ($F = 2.7$, $p = 0.046$ and $F = 5.2$, $p = 0.002$, respectively). Indeed, more variability in the active moiety plasma concentrations was explained by CYP2C19 metabolizer status than by CYP2D6 metabolizer status. Patients who were CYP2D6 PM/CYP2C19 NMs had 25% higher active moiety plasma concentrations compared to those who were CYP2D6 NM/CYP2C19 NMs.

Similarly, patients who were CYP2D6 IM/CYP2C19 IMs had 20% higher active moiety plasma concentrations compared to CYP2D6 NM/CYP2C19 NMs. Finally, the highest active moiety plasma concentration was observed in a CYP2D6 PM/CYP2C19 PM ($n = 1$) whose active moiety plasma concentration was 83% higher than CYP2D6 NM/CYP2C19 NMs. No other tested CYP450 enzyme was associated with concentrations of venlafaxine or total active moiety ($p \geq 0.05$).

Conclusions: Our results indicate that venlafaxine plasma concentrations were impacted significantly by CYP2C19, in addition to CYP2D6 metabolizer status. Given the known association between side effects and CYP2D6 metabolizer status, this suggests that both genes in combination may impact side effect risk. Furthermore, CYP2C19 was shown to be critical in associations with active moiety plasma concentrations. Future directions include assessing the impact of active moiety plasma concentrations and CYP2D6 in combination with CYP2C19 on venlafaxine efficacy and side effects. Pharmacogenetic guidelines for venlafaxine currently include therapeutic recommendations for CYP2D6 metabolizer status alone, but these results suggest that both CYP2D6 and CYP2C19 should be considered.

Keywords: pharmacogenomics, geriatric depression, Pharmacokinetics, Venlafaxine, desvenlafaxine

Disclosure: Myriad Genetics, Stock/Equity - Publicly Traded Company, Self, Myriad Genetics, Employee, Self.

P214. EXAMINING FRONTO-LIMBIC BRAIN AND SLEEP MECHANISMS OF ANTI-DEPRESSANT EFFECTS IN COGNITIVE-BEHAVIORAL THERAPY FOR INSOMNIA

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Background: Disturbed sleep, including insomnia, contributes to and maintain depression as well as disrupt emotion generation and regulation in fronto-limbic circuitry—by amplifying amygdala reactivity to negative stimuli while impairing top-down regulation by the medial prefrontal cortex (mPFC). Cognitive-Behavioral Therapy for Insomnia (CBT-I), the gold-standard insomnia treatment, reduces depressive symptoms, yet the neural and sleep mechanisms underlying these anti-depressant effects are unknown. Furthermore, while CBT-I consistently improves subjective insomnia symptoms, improvement in objective sleep is inconsistent, raising questions about whether subjective or objective sleep improvements drive its anti-depressant effects. Here we present primary outcomes from a single-arm first-phase of a two-phase mechanistic clinical trial with three aims: (1) whether CBT-I is associated with improvements in fronto-limbic brain function; (2) whether changes in fronto-limbic function are associated with changes in depressive symptoms and with improvements in both objective and subjective sleep disturbances, and whether changes in depressive symptoms are associated with changes in sleep disturbances; (3) whether baseline differences in fronto-limbic function and sleep can predict which participants experience the greatest anti-depressant benefit.

Methods: Forty-eight participants (64% female; age 25–59) with insomnia and depression symptoms completed the fMRI Facial Expressions of Emotion Task (supra- and subliminal conditions) and Emotion Regulation Scenes Task and had their depression

and insomnia symptoms assessed before and after receiving 6 weekly sessions of therapist-delivered CBT-I. Mechanistic outcomes were changes in amygdala reactivity and task-modulated connectivity between the amygdala and nodes of the mPFC. Depression was evaluated using the Beck Depression Inventory (BDI, excluding the sleep item), subjective insomnia symptoms using the Insomnia Severity Index (ISI), and sleep efficiency from polysomnography. The analysis plan was to first test for treatment-associated changes in amygdala reactivity using linear mixed-effects models with FDR correction applied for each task. If amygdala reactivity was significantly reduced, we then tested treatment-associated changes in connectivity between the amygdala and mPFC. Associations between fronto-limbic function and depression symptom improvement were examined using linear regression, as were associations between depression and insomnia symptom improvements. To test whether pre-treatment differences in fronto-limbic function, self-reported insomnia symptoms, and objective sleep efficiency were predictive of the magnitude of depression symptom improvement, we specified residualized change models, which isolate the relationships between pre-treatment inter-individual differences and post-treatment depression symptoms, accounting for baseline depression levels. All models adjusted for age and sex.

Results: Participants experienced a medium effect for reduced amygdala reactivity following CBT-I when viewing supraliminal fearful faces (Cohens $d = 0.55$, $b = -0.17$ [95% CI -0.27 , 0.06], p -adjusted = 0.008), and a small effect for increased amygdala connectivity with the sgACC ($d = 0.28$, $b = 0.16$ [0.008, 0.31], p -uncorrected = 0.045). No other significant treatment-related reductions in amygdala reactivity across all tasks (all p -adjusted ≥ 0.08) or increases in connectivity for supraliminal fearful faces were observed (all p -uncorrected ≥ 0.45).

Improved depression symptoms were not associated with either the reduction in amygdala reactivity to supraliminal fearful faces ($b = -2.9$ [-11.0 , 5.12], p -uncorrected = 0.47 , $\eta^2 p = 0.02$) or the increase in amygdala-sgACC connectivity ($b = -1.6$ [-7.1 , 3.8], p -uncorrected = 0.54 , $\eta^2 p = 0.01$). Instead, reduced depression symptoms were significantly associated with both increased objectively measured sleep efficiency ($b = -0.20$ [-0.39 , -0.01], p -uncorrected = 0.04 , $\eta^2 p = 0.10$) and reduced self-reported insomnia symptoms ($b = 0.90$ [0.37 , 1.4], p -uncorrected = 0.001 , $\eta^2 p = 0.19$).

Finally, the magnitude of reduced depressive symptoms was not predicted by pre-treatment amygdala reactivity ($b = 1.92$ [-5.8 , 9.6], p -uncorrected = 0.62 , $\eta^2 p = 0.003$) or sgACC connectivity ($b = 1.7$ [-0.4 , 8.5], p -uncorrected = 0.49 , $\eta^2 p = 0.04$). Only pre-treatment objective sleep efficiency, and not subjective insomnia symptoms, significantly predicted the reduction in depressive symptoms ($b = 0.21$ [0.06 , 0.36], p -uncorrected = 0.007 , $\eta^2 p = 0.16$), such that participants with the lowest objective sleep efficiency prior to treatment experienced greater improvements in depression symptoms.

Conclusions: This was the first study to assess neural mechanisms and predictors of CBT-I's anti-depressant effects. CBT-I was associated with improved fronto-limbic brain function, but these changes did not predict depression or insomnia improvements. While both objective and subjective insomnia symptoms improved and were associated with the magnitude of depression symptom improvement, only pre-treatment objective sleep efficiency predicted the subsequent magnitude of depression improvement, suggesting that those with highest objective sleep disturbance may experience the greatest anti-depressant benefit of CBT-I. These findings are being tested for replication in a large RCT as part of the study's second phase.

Keywords: Depression, Functional MRI (fMRI), polysomnograph, insomnia, CBT-I

Disclosure: Neurostar, Consultant, Self.

P215. MODERATING EFFECTS OF PSYCHIATRIC COMORBIDITIES, MEDICATIONS, AND DEMOGRAPHICS ON DEPRESSIVE SYMPTOM TRAJECTORIES DURING ACUTE-PHASE IV KETAMINE TREATMENT: AN ANALYSIS OF REAL-WORLD DATA

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Background: Major depressive disorder (MDD) and bipolar disorder (BP) are highly prevalent and debilitating psychiatric illnesses. MDD has a lifetime prevalence of about 12% while BD affects roughly 1% of the global population. Beyond their prevalence, both disorders impart high personal and societal costs including functional impairment and increased risk of suicide, underscoring the urgent need for rapidly acting and effective treatments. Despite the strong antidepressant effects observed in controlled trials, there remains a gap in knowledge regarding ketamine's real-world effectiveness and safety in routine practice. Most ketamine studies to date have been tightly controlled RCTs with modest sample sizes and short follow-up, conducted in academic or hospital settings. In recent years, off-label ketamine treatment has expanded into general outpatient practice, with ketamine infusion clinics and other services treating large numbers of patients outside of research studies. Systematic evaluation of outcomes in large, unselected patient cohorts is necessary. This study examines the moderating effects of BP, baseline psychiatric comorbidities, and concurrent medication use on depressive symptom trajectories during acute-phase IV ketamine treatment in a large naturalistic outpatient cohort.

Methods: Adult patients (age = 41.4 [SD = 13.6]; 56% women) with MDD ($n = 1611$) or BD ($n = 188$) received IV ketamine in an outpatient network with high adherence to a recommended protocol of four infusions over 14 days ($n = 1565$) while others ($n = 234$) were treated off-protocol or discontinued treatment. Depressive symptoms were assessed with the PHQ-9 which was acquired within 30 days pre-treatment, at visits #1 and #4, and +3, +9, and +14 days post-treatment. At intake, patients self-reported psychiatric comorbidities including anxiety ($n = 1304$), obsessive-compulsive disorder (OCD; $n = 222$), post-traumatic stress disorder (PTSD; $n = 502$), substance use disorder (SUD; $n = 180$), and suicidal ideation (SI; $n = 1024$) and concurrent medication use. Medications were categorized as antidepressants ($n = 1117$), antipsychotics ($n = 298$), benzodiazepines ($n = 462$), non-benzodiazepine anxiolytic sedatives (NBAS; $n = 275$), mood stabilizers ($n = 321$), stimulants ($n = 393$), and other psychotropic medications ($n = 239$). Linear mixed-effects models (LMMs) characterized symptom trajectories over log-transformed time (days) and tested moderating effects of psychiatric diagnoses and concurrent medication use, adjusting for baseline symptoms, age, and sex. Secondary analyses examined moderating effects of age (adult [ages 18–64] versus senior [ages 65 and older]) and sex. Patients were included as random intercepts. Multiple comparisons were adjusted using the false discovery rate.

Results: The LMM included 8965 repeated PHQ-9 measures on 1799 unique patients and explained substantial variance (R^2 marginal = 0.51 and R^2 conditional = 0.73). PHQ-9 scores decreased significantly over time ($\beta = -1.66$ [95% CI: -1.70 , -1.62], $q < 0.001$). Patients with a primary diagnosis of BD showed accelerated improvement compared to those with MDD ($\beta = -0.16$ [95% CI: -0.28 , -0.04], $q < 0.05$). Among self-reported comorbidities, patients with PTSD ($\beta = -0.18$ [95% CI: -0.27 , -0.10], $q < 0.001$), SUD ($\beta = -0.21$ [95% CI: -0.34 , -0.08], $q < 0.01$), and SI ($\beta = -0.43$ [95% CI: -0.51 , -0.36], $q < 0.001$) experienced accelerated symptom improvement while those with

OCD had a more blunted recovery ($\beta = 0.20$ [95% CI: 0.08, 0.31], $q < 0.01$). No significant effects of anxiety were observed. No time-varying effects of medications were observed to moderate the rate of symptom trajectories, however main effects of benzodiazepines ($\beta = 0.62$ [95% CI: 0.22, 1.03], $q < 0.01$) and NBAS ($\beta = 0.59$ [95% CI: 0.11, 1.06], $q < 0.05$) were observed. Secondary analyses revealed that seniors experienced a more protracted rate of recovery relative to adults ($\beta = 0.48$ [95% CI: 0.33, 0.62], $q < 0.001$) and males similarly had a slower recovery relative to women ($\beta = 0.14$ [95% CI: 0.06, 0.21], $q < 0.01$).

Conclusions: In this large, naturalistic outpatient cohort, repeated IV ketamine infusions were associated with robust improvements in depressive symptoms, corroborating treatment efficacy observed in controlled trials into routine practice. Patient diagnosis, comorbidity, and demographic factors moderated symptom trajectories. Specifically, patients with BD, PTSD, SUD, or baseline suicidal ideation showed accelerated improvement, while those with OCD, older age, or males experienced slower recovery. Concomitant use of benzodiazepines and non-benzodiazepine sedatives was associated with higher symptom burden; however, no medication class altered the rate of symptom change treatment course. These findings suggest that ketamine's antidepressant effects generalize to real-world patients with complex comorbidities, while highlighting specific subgroups who may require further monitoring or adjunctive treatment strategies. Systematic collection of outcomes across clinical settings will be critical to informing patient selection for ketamine treatment.

Keywords: Depression, Bipolar Disorder, IV- Ketamine, Psychiatric Comorbidity, longitudinal studies

Disclosure: Nothing to disclose.

P216. IN UTERO SSRI EXPOSURE IS ASSOCIATED WITH ALTERED RESTING-STATE ACTIVITY IN YOUTH CONSISTENT ACROSS TWO TIME POINTS

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Background: Maternal anxiety and depression during pregnancy are associated with adverse child outcomes including altered neurodevelopment and increased risk for mood and anxiety disorders. Around ~6% of pregnant mothers in the US are taking selective serotonin reuptake inhibitors (SSRIs) to combat anxiety and depression. SSRIs readily cross the placenta and may thus affect fetal development. Serotonin is a crucial neurotransmitter during development, with functions ranging from cell proliferation, differentiation, synaptogenesis, and migration. Alterations in gestational serotonin via SSRI exposure have been linked to adverse neuroanatomical and behavioral outcomes. However, the effects of in SSRI exposure on child development remain largely unclear and need to be weighed against the alternative risks of exposure to maternal depression.

In mice and human youth, we recently showed heightened anxiety and depressive behaviors and increased activity in the fear circuit during innate fear tasks. Building on this prior work, here we assess the effects of in utero SSRI exposure on fMRI resting state activity in human youth.

Methods: We used the large and diverse Adolescent Brain and Cognitive Development Study (ABCD; analytic sample $N = 4550$; $n = 107$ SSRI exposed in utero; $n = 2343$ boys, $n = 2198$ girls). Mothers retrospectively reported on SSRI use and other medications and substances administered during pregnancy which were

controlled for in the statistical analyses, including smoking, illicit drugs, alcohol and caffeine. Analyses included propensity score weighting to control for potential confounding of who takes SSRIs in pregnancy and who does not (including maternal race/ethnicity, mother's age at pregnancy, planned pregnancy and substance use). Children underwent MRI scans every two years. Each session included four 5 min resting state runs (passive viewing of a cross-hair). The outcome measures for the current project are fMRI resting state correlations between five Gordon networks (fronto-opercular, default mode, salience, dorsal attention, frontoparietal) and the amygdala and hippocampus at age 9/10 (baseline) and 11/12 (two-year follow up). We chose these two regions as we found in our previous work that they were most impacted in in utero SSRI exposed compared to unexposed groups. We then determined whether network activity was associated with future depressive symptoms (child behavior checklist DSM depressive symptoms) in the youth. Linear mixed models (lme4 in RStudio) accounted for family structure and imaging site, with adjustments for sex, age, maternal depression and symptomatology, and issues related to pregnancy and birth (such as pregnancy complications, vitamin use, birth weight, doctors' visits, birth by caesarian). P-values were adjusted for multiple comparisons using false discovery rate (FDR) correction.

Results: Consistently at both age 9/10 and age 11/12 there was increased activity between the amygdala and the fronto-opercular network (bilateral; standardized betas ~0.3, FDR-adjusted ps: 0.01–0.00001) and the left amygdala and the salience network (standardized betas 0.15–0.25; FDR-adjusted ps: 0.05–0.007), while there was decreased activity between the right amygdala and salience network and between the left hippocampus and the salience network (standardized betas 0.15–0.25, FDR corrected ps 0.01–0.0001) in youth exposed to SSRIs in utero compared to controls. Increased activity in the left amygdala-salience network and decreased activity in the right amygdala-salience and left hippocampal-salience network at baseline was associated with future depressive symptoms in the youth two years later (st.betas 0.03–0.09, FDR-corrected ps < 0.05). These differences were observed while controlling for maternal lifetime depression and maternal symptomatology at time of MRI scan. When comparing in utero SSRI exposure within the subgroup of offspring of mothers with lifetime depression, the differences in the amygdala-opercular network and left hippocampal-salience network remained present.

Conclusions: Children exposed to SSRIs in utero had highly significant differences in resting state brain activity related to integrating emotion (regulation) with cognitive control and filtering salient stimuli, and these differences in brain activity were associated with future depressive symptoms. Our findings suggest SSRI use during pregnancy may have adverse effects for the offspring. As maternal depression during pregnancy may also lead to increased depression and altered neurodevelopment in the offspring, our findings indicate further research is needed to understand whether other treatments, such as non-SSRI antidepressants and psychotherapy, might be preferable to SSRIs in order to decrease depression during pregnancy.

Keywords: in utero SSRI exposure, prenatal depression, Resting State Functional Connectivity, youth depression

Disclosure: Nothing to disclose.

P217. SUICIDAL IDEATION AND BEHAVIOR IN PATIENTS WITH TREATMENT-RESISTANT DEPRESSION TREATED WITH GH001

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Background: Treatment-resistant depression (TRD) is associated with reduced quality of life, impairment in psychosocial function, and higher burden of treatment compared with treatment-responsive major depressive disorder. Patients with TRD also experience a disproportionately higher rate of suicide. Thus, there is a significant unmet need for safe and effective therapies for TRD that do not exacerbate suicidal intent. In a Phase 2b trial, GH001, a synthetic form of mebufotenin (5-methoxy-N,N-dimethyltryptamine [5-MeO-DMT]) for pulmonary inhalation, was well tolerated and resulted in rapid and significant improvements in depressive symptoms. Here we describe the effects of GH001 on suicidal ideation and behavior in patients with TRD enrolled in the Phase 2b trial.

Methods: This trial (NCT05800860) included a 7-day, randomized, double-blind part (Part 1) and a 6-month open-label extension (OLE; Part 2). In Part 1, patients were randomized 1:1 to receive an individualized dosing regimen (IDR) of up to three escalating doses of GH001 (6, 12, and 18 mg) or placebo IDR on a single day with a 1-h interval between doses. The criteria for administration of the second and third doses in the IDR were based on patients' subjectively reported psychoactive effects, and the safety and tolerability at the previous dose level(s), according to the trial physician's judgement. In Part 2, patients could receive up to five GH001 IDR re-treatments over 6 months as needed; criteria for re-treatment were based on Montgomery-Åsberg Depression Rating Scale (MADRS) score and safety and tolerability of the previous dose(s). Trial eligibility criteria excluded patients with suicidal ideation with intent (with items 4 or 5 on the Columbia-Suicide Severity Rating Scale [C-SSRS] endorsed) within the past year or at screening or baseline; those with suicidal behaviors or non-suicidal self-injury in the past year; and those with a clinical assessment of significant suicidal risk. C-SSRS scores were assessed at all visits during Parts 1 and 2 of the trial. The clinician-rated C-SSRS quantifies suicidal ideation and behavior through a semi-structured interview. In addition, MADRS item 10 provided further quantification of suicidal ideation at frequent timepoints throughout the trial. Results were analyzed descriptively.

Results: Eighty-one patients enrolled in the trial; 40 were randomized to GH001 IDR and 41 to placebo IDR in Part 1, and all transitioned directly into Part 2. Mean (SD) patient age was 42.8 (11.2) years, and 56.8% of patients were female. The number of patients with historic and current suicidal ideation were represented evenly across the GH001 and placebo groups; at screening, 21 patients (GH001, $n = 10$; placebo, $n = 11$) reported a lifetime history of suicidal ideation and 19 patients (GH001, $n = 8$; placebo, $n = 11$) reported suicidal ideation during the 12 months before screening, and at baseline, 14 patients reported current suicidal ideation via C-SSRS assessment (GH001, $n = 7$; placebo, $n = 7$). During Part 1, at Day 8, suicidal ideation without intent was reported for four patients in the GH001 group (all of whom had reported it at baseline) and seven patients in the placebo group (5/7 reported suicidal ideation at baseline). In Part 2 of the trial, the numbers of patients reporting suicidal ideation at all timepoints assessed during the trial were lower than that at baseline. The median (range) MADRS item 10 score was 0.0 (0 to 4) at baseline and the same at 6 months; median change from baseline (range) was 0.0 (−3 to 2) at 6 months. Across both trial parts, there were no treatment-emergent events of self-harm or suicidal behavior or of suicidal ideation with intent during follow-up. There was one treatment-emergent adverse event (TEAE) of suicidal ideation during Part 2 that was assessed as moderate in intensity, not

classified as serious, and possibly related to GH001; this event lasted for 6 hours before resolving spontaneously. This TEAE was not accompanied by any changes in C-SSRS classification beyond the duration for which the thoughts occurred, and the patient did not report any further TEAEs of suicidal ideation during the trial.

Conclusions: In this double-blind, placebo-controlled trial with a 6-month OLE in patients with TRD, GH001 was not associated with treatment-emergent events of suicidal ideation with intent or behavior. These findings indicate that GH001 could potentially provide significant reductions in depressive symptoms without increasing the risk of suicide in patients with TRD.

Keywords: Treatment-Resistant Depression, Psychedelics, Suicidality

Disclosure: Autobahn, Consultant, Self, Boehringer-Ingelheim, Consultant, Self, Clexio Biosciences, Consultant, Self, Engrail Therapeutics, Consultant, Self, EMA Wellness, Consultant, Self, Engrail Therapeutics, Consultant, Self, Motif Neurotech, Consultant, Self, Neumora, Consultant, Self, Newleos, Consultant, Self, Relmada Therapeutics, Consultant, Self, Signant Health, Consultant, Self, Supernus, Consultant, Self, Xenon, Consultant, Self.

P218. PEER CHRONIC STRESS, NEURAL RESPONSE TO PEER FEEDBACK, AND DEPRESSION SYMPTOMS IN ADOLESCENT GIRLS

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Background: Interpersonal chronic stress, particularly with peers, is a robust predictor of depression risk in adolescent girls. However, not all youth exposed to this stress become depressed, raising questions about which adolescents are most susceptible. One potential risk factor that may modulate vulnerability to peer stress is heightened subgenual anterior cingulate cortex (sgACC) reactivity to peer rejection. The sgACC is involved in processing social feedback, and activation of this region during peer exclusion correlates with concurrent emotional distress. Furthermore, greater sgACC reactivity to peer rejection or exclusion has been shown to predict adolescent depression risk both cross-sectionally and prospectively. Yet, it remains unclear whether increased sgACC response to peer rejection also exacerbates the impact of real-world peer stress on depression risk. In this preliminary study, we tested this question in a risk-enhanced sample of adolescent girls by examining the interactive effect of peer chronic stress exposure and sgACC reactivity to peer feedback on depression symptom severity.

Methods: This sample included 32 female adolescents at-risk for depression (i.e., with a parental history of major depressive disorder) enrolled in an ongoing study. Youth were 13 to 15 years old (Mean = 14.57, SD = 0.89), 59.4% identified as White, and 31.3% identified as Latine. Youth completed the Mood and Feelings Questionnaire to assess current depression symptoms and were administered the Youth Life Stress Interview to assess interpersonal chronic stress within multiple domains (i.e., Peer, Romantic, Family). Youth also completed the Chatroom Interact Task during functional magnetic resonance imaging to assess neural response to peer feedback. In this task, youth were "matched" with two "peers" and were told they would take turns choosing who they would prefer to talk to about various topics. Task conditions include "Acceptance" trials (in which they were chosen), "Rejection" trials (in which they were not chosen), and a motor "Control" condition. Neural activation (β weights) was extracted for sgACC for the Acceptance > Control and Rejection >

Control contrasts for analysis. Linear regression analyses were conducted to examine the main and interactive effects of sgACC reactivity (Acceptance > Control or Rejection > Control) and interpersonal chronic stress (peer, romantic, or family) on depression symptoms.

Results: Preliminary results indicated that both the Peer Stress x sgACC response to rejection, $\beta = 2.58$, $t(27) = 2.48$, $p = 0.020$, and the Peer Stress x sgACC response to acceptance, $\beta = 2.97$, $t(27) = 2.42$, $p = 0.023$, interactions were significant. Simple slopes for the Peer Stress x sgACC to rejection interaction indicated that peer chronic stress was positively associated with depression symptoms for youth exhibiting greater (+1 SD) sgACC response, $\beta = 0.71$, $t(27) = 2.38$, $p = 0.025$, but not lower (−1 SD) sgACC response, $\beta = -0.14$, $t(27) = -0.66$, $p = 0.516$, to rejection feedback. Similar effects were observed for the interaction with sgACC to acceptance, such that peer chronic stress was positively associated with depression symptoms for youth exhibiting greater (+1 SD) sgACC response, $\beta = 0.779$, $t(27) = 2.42$, $p = 0.023$, but not lower (−1 SD) sgACC response, $\beta = -0.14$, $t(27) = -0.68$, $p = 0.500$, to acceptance feedback. No other main or interactive effects of sgACC or interpersonal stress were significant (lowest $p = 0.192$).

Conclusions: Preliminary findings suggest that peer chronic stress is associated with greater depression symptoms, specifically for adolescent girls who exhibit heightened sgACC response to peer feedback. Unexpectedly, this was observed for both rejection and acceptance feedback, suggesting that increased reactivity to peer feedback more broadly may increase adolescent susceptibility to peer stress. This study is ongoing, and results are preliminary. Once data collection is complete, future work will examine whether sgACC response to peer feedback interacts with peer stress to prospectively predict depression symptom increases over a multi-wave follow-up.

Keywords: Functional MRI (fMRI), Brain Based Markers for Depression, affective neuroscience, Peer Stress

Disclosure: Nothing to disclose.

P219. EFFICACY OF ADJUNCTIVE LUMATEPERONE 42 MG TREATMENT ACROSS DEPRESSION AND ANHEDONIA SYMPTOMS IN MAJOR DEPRESSIVE DISORDER

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Background: Current treatments for major depressive disorder (MDD) may not resolve all symptoms, with depressed mood and anhedonia often persisting with treatment. In people with MDD residual symptoms predict relapse, recurrence, and functional impairment [1]. Lumateperone is a mechanistically novel FDA-approved antipsychotic to treat schizophrenia and depressive episodes associated with bipolar I or bipolar II disorder [2]. A recent positive Phase 3, randomized, double-blind, placebo-controlled, multicenter trial (Study 502, NCT05061706), investigated adjunctive lumateperone 42 mg in patients with MDD with inadequate response to ongoing antidepressant therapy (ADT). This analysis of Study 502 assessed the efficacy of lumateperone across depression symptoms measured using Montgomery-Asberg Depression Rating Scale (MADRS) single-item scores and the MADRS anhedonia factor.

Methods: Eligible adults (18–65 years) met DSM-5 criteria for MDD with inadequate response to 1 or 2 ADT in the current depressive episode (<50% improvement on the Antidepressant Treatment Response Questionnaire) and had MADRS Total score

≥24, Clinical Global Impression Scale-Severity score ≥4, and Quick Inventory of Depressive Symptomatology-Self Report-16 item score ≥14. Patients were randomized to 6-weeks of oral placebo or lumateperone 42 mg adjunctive to ADT. A prospective analysis measured change in individual MADRS items using a mixed-effects model for repeated measures (MMRM) in the modified intent-to-treat (mITT) population. A post-hoc analysis evaluated anhedonia symptoms according to MADRS anhedonia factor (sum of apparent sadness, reported sadness, concentration difficulties, lassitude, and inability to feel) using an MMRM in the mITT.

Results: Of 480 patients treated, 469 were included in the mITT population (lumateperone+ADT, 232; placebo+ADT, 237) and 429 (89.4%) completed treatment. Adjunctive lumateperone 42 mg met the primary endpoint, significantly improving depression symptoms at Day 43 according to MADRS Total score compared with adjunctive placebo (least-squares mean difference vs adjunctive placebo [LSMD], −4.5; ES, −0.56; $P < 0.0001$). The most prominent MADRS items at baseline were apparent sadness (mean score: lumateperone+ADT, 3.8; placebo+ADT, 3.9) and reported sadness (lumateperone+ADT, 4.2; placebo, 4.2). At Day 43, all 10 MADRS items significantly improved with adjunctive lumateperone compared with adjunctive placebo: apparent sadness (LSMD, −0.6; $P < 0.0001$), reported sadness (LSMD, −0.7; $P < 0.0001$), inner tension (LSMD, −0.4; $P < 0.001$), reduced sleep (LSMD, −1.0; $P < 0.0001$), reduced appetite (LSMD, −0.4; $P < 0.001$), concentration difficulties (LSMD, −0.4; $P < 0.001$), lassitude (LSMD, −0.5; $P < 0.001$), inability to feel (LSMD, −0.4; $P < 0.001$), pessimistic thoughts (LSMD, −0.6; $P < 0.0001$), and suicidal thoughts (LSMD, −0.1; $P < 0.05$). Improvements with lumateperone+ADT were significant at every visit for reduced sleep and improvements were significant at Day 15 onward for apparent sadness and reported sadness.

Lumateperone+ADT significantly improved anhedonia symptoms compared with placebo+ADT at Day 22 onward measured by MADRS anhedonia factor score (Day 43 LSMD, −2.4; ES, −0.47; $P < 0.0001$). Each of the items included in the MADRS anhedonia factor significantly improved with adjunctive lumateperone by Day 29.

Conclusions: Adjunctive lumateperone 42 mg to ADT demonstrated efficacy, significantly improving a broad range of depression symptoms, including anhedonia, compared with adjunctive placebo in patients with MDD with inadequate response to ADT.

Keywords: Major Depressive Disorder (MDD), Reward, motivation and Anhedonia, lumateperone

Disclosure: Abbvie, Consultant, Self, Alkermes, Consultant, Self, Alkermes, Speakers Bureau, Self, Alkermes, Consultant, Self, American Psychiatric Publishing, Royalties, Self, Axsome, Speakers Bureau, Self, Bristol Myers Squibb, Consultant, Self, Bristol Myers Squibb, Speakers Bureau, Self, Cambridge Univ Press, Royalties, Self, Alvogen, Consultant, Self, Genomind, Consultant, Self, Luye Pharmaceuticals, Advisory Board, Self, Neumora, Advisory Board, Self, Otsuka, Consultant, Self, Otsuka, Honoraria, Self, Intracellular Therapies, Speakers Bureau, Self, Intracellular Therapies, Consultant, Self, Vanda Pharmaceuticals, Speakers Bureau, Self.

P220. SUBSTANCE USE, ANHEDONIA, AND NEURAL REWARD FUNCTION IN ADOLESCENTS AT RISK FOR PSYCHOPATHOLOGY

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Background: Adolescence is a vulnerable developmental period in which youth are susceptible to developing psychopathology. Anhedonia—referred to as diminished experiences of pleasure—

often emerges in adolescence, predicts psychopathology such as depression and substance use disorders, and is associated with disruptions in neural reward circuitry. Early substance use is also a known predictor of later psychopathology and may be especially reinforcing for people with anhedonia due to its rewarding neuropsychological properties. Studying earlier developmental stages in youth, prior to the onset of psychopathology, may reveal important insights regarding mechanisms and clinical correlates.

Methods: 117 adolescents at low/high familial risk for developing psychopathology (major depression, bipolar disorder, schizophrenia/schizoaffective disorders) ages 13–18 years (55% girls) completed clinical measures and fMRI scanning during a guessing reward task. Functional neuroimaging data were collected using a 3.0 Tesla Siemens Prisma MRI scanner. Preprocessing and fMRI analyses were performed using SPM. ROIs included the ventral striatum (VS) and the medial prefrontal cortex (mPFC), key regions of reward processing. Logistic moderation models tested whether anhedonia and neural reward function of the VS and mPFC interacted to predict history of substance use.

Results: Logistic regression revealed a significant interaction between anhedonia and right VS (but not mPFC) activation (win outcome > loss outcome) in predicting prior substance use (Int: $b = -0.18$, $SE = 0.07$, $p = 0.013$). Johnson–Neyman analysis indicated at low levels of right VS activation, higher anhedonia severity was associated with increased odds of substance use ($b = 0.27$, $SE = 0.10$, $p = 0.007$), whereas at average levels of right VS activation, the effect was smaller ($b = 0.11$, $SE = .06$, $p = 0.071$), and at high levels, the effect reversed ($b = -0.11$, $SE = 0.09$, $p = 0.224$). Further probing of the interaction revealed that VS activation was positively associated with the probability of substance use among youth with low anhedonia, but this association weakened as anhedonia severity increased. For youth with more anhedonia severity, the likelihood of prior substance use was elevated regardless of VS activation. Findings held when controlling for sex.

Conclusions: These findings indicate the possible presence of distinct pathways associated with adolescent substance use history. Among youth with low anhedonia, higher VS responsiveness to reward is linked to increased substance use, suggesting a relationship with heightened reward sensitivity. Conversely, in youth with more anhedonia severity, substance use history is elevated regardless of neural reward activation. These results underscore the value of considering both subjective affective experience and neural reward functioning when studying substance use behaviors in adolescents.

Keywords: anhedonia, Monetary reward, Substance abuse

Disclosure: Nothing to disclose.

P221. NEUROCOGNITIVE COMPONENTS OF NEGATIVE SELF-ASSOCIATIONS IN A DEPRESSED SAMPLE

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Background: Clinical and phenomenological studies have pinpointed that depression deeply affects the experience of self. Mental associations between one's self-concept and negative attributes are hypothesized to play a central role in the etiology and maintenance of depression. Although the effects of depression on experience of self are well documented, how this mostly self-reported and phenomenologically studied phenomenon is manifested in other levels of analysis has been less studied. Here we used a cognitive task, the Implicit Association Test (IAT), which was completed inside an fMRI scanner and a computational framework for analyzing the performance on the task to study

neurocognitive components of negative self-associations in depression.

The IAT is a simple task which is thought to reflect representations in memory that link a stimulus and an involuntarily activated evaluative outcome which do not require conscious reflection. The IAT measures implicit associations based on the relative speed with which an individual classifies words (e.g., "worthless" and "me"). Performance on IAT can be formalized using Drift Diffusion Models (DDM) that have been widely applied in speeded decision-making tasks in cognitive psychology to yield insights into the computational processes that underlie decisions on a trial-by-trial bases. We used DDMs with three main parameters: drift rate which represents the quality of integrating stimulus information, boundary separation which indexes a participant's general tendency toward cautious, and nondesired time which represents the latency of the processes outside the decision making itself.

In the current study, treatment-seeking adults with moderate-to-severe, treatment-resistant depression completed a novel battery of 4 IATs (assessing self-associations) inside an fMRI scanner. We aimed to: (1) assess the relationship between the computational substrates of IAT performance and relevant clinical self-report (explicit) measures; and (2) assess neural substrates of IAT performance, to yield novel insights regarding the nature and mechanisms of self-associations in the context of clinical depression.

Methods: $N = 155$ depressed patients (62% female) between the ages of 18–60 with moderate-to-severe levels of depression [Montgomery-Asberg Depression Rating Scale (MADRS) score ≥ 25], lower-than-normative self-reported self-esteem/self-worth [based on their scores on the Cognitive Triad Inventory "self" subscale and/or the Rosenberg Self-Esteem Scale (RSES)], and at least one failed, adequate trial of an FDA-approved antidepressant medication within the current depressive episode, were recruited for an experimental intervention study and completed a baseline study visit. A battery of four different types of IAT were completed within a 3Tesla Siemens PRISMA scanner where word stimuli assessing the relationship between self and depression-related constructs ('bad,' 'worthless,' 'sad,' and 'escape') were presented.

We used a HDDM (hierarchical drift diffusion model) toolbox to estimate the parameters of DDMs during a battery of IATs designed to capture self-referential associations. Hierarchical Bayesian parameter estimation methods are useful for enhancing statistical power, allowing for simultaneous estimation of individual subject parameters and the group distribution that they are drawn from.

Since we were interested in the neural substrates of the relative response time when self and positive attributes were paired compared to when self and negative attributes were paired, after standard preprocessing we subtracted the beta weights of the two critical blocks and then used AFNI's 3dRegAna to find the association between the difference beta weight maps and the computationally computed parameters from the 4 word-based IATs. To correct for multiple comparisons across the whole brain, we employed Threshold-Free Cluster Enhancement (TFCE) as implemented in FSL's randomise tool with $N = 5000$ and family-wise error (FWE) correction was applied at $p < 0.05$.

Results: All models showed good convergence as evident by trace plots and the Gelman-Rubin $\hat{R}$ statistic. We observed significant correlations between drift rate and RSES for the IATs assessing implicit self-esteem [self-association with the constructs 'bad' and 'worthless' ($r(149) = -0.26$, $p < 0.01$)]. We didn't observe a significant relationship between the computational parameters of the IAT assessing self-associations with sadness and clinician-rated depression severity (MADRS total scores), or between the IAT assessing self-associations with 'escape' and suicidal ideations measured by the Columbia Suicide Severity Rating Scale (CSSRS). In neuroimaging analyses, we observed significant clusters in

middle frontal gyrus, superior frontal gyrus, frontal pole and precuneus regions where neural activation was associated with the overall drift rate across all IATS (map-wise TFCE p 's < 0.05).

Conclusions: Our behavioral, neural, and computational results suggest IAT performance in moderate-to-severe, treatment-resistant depress patients is characterized by overall increased demands on cognitive neural resources, and less efficient information accumulation, when positive self-attributes are displayed. In the context of depression and low explicit self-esteem, self-referential processing of positive attributes is both less efficient and more neurocognitively demanding.

Keywords: drift diffusion model, computational models of decision-making, Implicit association test, Depression

Disclosure: Nothing to disclose.

P222. DIFFERENTIAL ASSOCIATIONS OF BODY MASS INDEX WITH DEPRESSION, ANXIETY, AND PTSD SYMPTOM TRAJECTORIES IN PATIENTS RECEIVING KETAMINE INFUSION THERAPY IN THE REAL WORLD

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Background: Ketamine infusion therapy (KIT) is increasingly used to treat treatment-resistant depression, anxiety, and posttraumatic stress disorder (PTSD). Identifying clinical factors associated with symptom trajectories during KIT can inform personalized treatment. Body mass index (BMI), an indicator of metabolic health, is linked to inflammatory processes that may impact psychiatric outcomes (e.g., Morris and Vaccarino, 2013). Some studies have suggested that elevated BMI may predict a more robust antidepressant response to KIT (Freeman et al., 2020; Tan et al., 2025), though these findings remain inconsistent. Additionally, it is not yet clear whether similar patterns hold for other psychiatric outcomes in real-world clinical settings. As such, this study investigates linear and quadratic associations of BMI with average symptom trajectories of depression, anxiety, and PTSD symptoms in a real-world outpatient cohort receiving KIT. We also explored whether BMI moderates symptom change over treatment by testing interactions with the number of infusions received.

Methods: Repeated measures of depression (PHQ-9), anxiety (GAD-7), and PTSD (PCL-5) symptoms were collected from patients receiving KIT in practices across the US, recorded in the Osmind EHR system. The analytic samples differed by outcome and diagnoses, including only patients with a clinical diagnosis of the respective disorder and available questionnaire data: $N = 10,354$ patients for the PHQ-9 model, $N = 4536$ for the GAD-7 model, and $N = 1600$ for the PCL-5 model. For each outcome, linear mixed effects models were initially fit including fixed effects for BMI (z-scored), number of treatments (log-transformed), age, sex, race, dose, treatment density, and random intercepts and slopes by patient. Quadratic BMI terms were subsequently added to assess potential nonlinear relationships. Interaction terms between BMI and number of treatments were also tested to examine whether BMI moderated symptom trajectories.

Results: The distribution of patients across BMI categories was similar across diagnostic cohorts: underweight patients were the smallest group and roughly one-third were normal weight or over weight. Notably, 28.1% of the MDD sample, 26.1% of the GAD sample, and 38.9% of the PTSD sample were classified within the Obese categories. In the mixed effects models, linear BMI terms revealed a significant negative association with anxiety symptoms ($b = -0.19$, $p < 0.01$), and with PTSD symptoms ($b = -0.93$, $p < 0.001$), with higher BMI linked to lower average symptom

levels across sessions. For depression, linear BMI was not significant on its own. However, the linear term became significant with the addition of the quadratic term ($b = -0.17$, $p < 0.05$). The quadratic term also showed a significant positive association with depressive symptoms ($b = 0.11$, $p < 0.001$), indicating a U-shaped relationship in which both low and high BMI were associated with higher average PHQ-9 scores over time.

In models testing interaction effects, the BMI-by-treatment number interaction was significant only for PHQ-9 outcomes ($b = -0.10$, $p < 0.05$), suggesting that individuals with higher BMI experienced greater reductions in depressive symptoms over the course of treatment. The interaction was not significant for PTSD or anxiety symptoms. Sensitivity analyses were conducted by categorizing patients into those more likely versus less likely to have metabolic dysregulation, specifically comparing Class II/III obesity ($BMI > 35$) to all other BMI categories. Results were consistent with the primary analyses, supporting that observed effects were not solely driven by BMI magnitude but also reflect groups at higher metabolic risk.

Conclusions: Using real-world data from the Osmind EHR system, this study highlights a complex relationship between BMI and outcomes for KIT across psychiatric disorders, with distinct patterns observed for depression, anxiety, and PTSD. Higher BMI was associated with lower average anxiety and PTSD symptoms during treatment, while depression exhibited a nonlinear U-shaped relationship, with both low and high BMI linked to greater symptom burden. Importantly, BMI moderated response to ketamine for depression, where patients with higher BMI showed slightly greater symptom improvement across treatments. These findings, drawn from one of the largest real-world KIT cohorts to date, suggest that metabolic health (as reflected by BMI) may shape both average symptom severity and treatment responsiveness, with the latter particularly for depression, highlighting the potential for tailoring ketamine therapy based on patient clinical profiles.

Keywords: ketamine, metabolic, PTSD, Depression, Anxiety

Disclosure: Nothing to disclose.

P223. SHORT- AND LONG-TERM EFFECTS OF ESKETAMINE NASAL SPRAY ON ANHEDONIA IN TREATMENT-RESISTANT DEPRESSION: POST-HOC ANALYSES FROM TWO PHASE 3 STUDIES

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Background: Approximately 30% of patients with major depressive disorder (MDD) do not achieve remission despite treatment with multiple antidepressants, and those who do respond experience delayed onset of effect, leaving many impacted by the associated harm of core MDD symptoms such as anhedonia. Anhedonia is diminished interest and/or pleasure in usual activities and is linked to poor MDD prognosis. There is a critical need for novel treatments targeting the underlying MDD pathophysiologic pathways to provide rapid relief, especially in treatment-resistant depression (TRD). Esketamine nasal spray, a glutamate modulator and the S-enantiomer of racemic ketamine, was initially approved for TRD alongside an oral antidepressant (OAD) and has recently been approved as a standalone monotherapy treatment. Findings from the Phase 3, 4-week, double-blind (DB), TRANSFORM-2 study (NCT02418585) that supported esketamine's initial approval demonstrated significant and clinically meaningful reductions in Montgomery  sberg Depression Rating Scale (MADRS) total score with esketamine

plus OAD compared with placebo plus OAD as early as 24 h post first dose. Here we report post-hoc analyses of the short- and long-term effects of esketamine on anhedonia from TRANSFORM-2 and from the Phase 3, 52-week, open-label, SUSTAIN-2 (NCT02497287) study.

Methods: Adults with MDD and nonresponse to ≥ 2 OADs in the current episode were eligible for enrollment in TRANSFORM-2 and SUSTAIN-2. TRANSFORM-2 participants were randomized 1:1 to receive flexibly dosed esketamine (56 or 84 mg) or placebo twice/week for 4 weeks (induction treatment), both plus a new OAD. SUSTAIN-2 participants were treated with a 4-week induction phase of esketamine 28 mg (≥ 65 years old), 56 mg, or 84 mg twice/week followed by a 48-week open-label optimization/maintenance (O/M) phase in which esketamine dosing frequency was either once/per week (weeks 5–8 and thereafter) or every other week, plus a new OAD. Changes from baseline in MADRS anhedonia factor score (range: 0–30) and in the Patient Health Questionnaire-9 (PHQ-9) item 1 (little interest or pleasure in doing things) score (range: 0–3) were assessed; mean changes between placebo+OAD and esketamine+OAD were evaluated using analysis of covariance models for TRANSFORM-2. The percentage of participants achieving $\geq 50\%$ reduction in MADRS anhedonia factor score over time was reported; comparison between placebo+OAD and esketamine+OAD was conducted via Cochran-Mantel-Haenszel test for TRANSFORM-2. All p values are nominal.

Results: In TRANSFORM-2, the mean (SD) MADRS anhedonia factor score at baseline was 22.1 (2.83; $n = 109$) for placebo+OAD and 21.5 (2.97; $n = 114$) for esketamine+OAD; mean (SD) change from baseline to day 2 was -3.5 (5.61; $n = 102$) for placebo+OAD and -5.3 (5.86; $n = 109$) for esketamine+OAD ($p = 0.004$) and from baseline to day 28 was -10.3 (7.95; $n = 100$) for placebo+OAD and -12.6 (7.30; $n = 101$) for esketamine+OAD ($p = 0.002$). The percentage of participants achieving $\geq 50\%$ reduction in MADRS anhedonia factor score at day 2 was 10.8% (11/102) for placebo+OAD and 17.4% (19/109) for esketamine+OAD ($p = 0.158$) and at day 28 was 52.0% (52/100) for placebo+OAD and 69.3% (70/101) for esketamine+OAD ($p = 0.007$). The mean (SD) PHQ-9 item 1 score at baseline was 2.7 (0.55; $n = 109$) for placebo+OAD and 2.7 (0.56; $n = 114$) for esketamine+OAD; mean (SD) change from baseline to day 28 was -1.3 (1.05; $n = 99$) for placebo+OAD and -1.7 (1.02; $n = 101$) for esketamine+OAD ($p = 0.006$). In SUSTAIN-2, the mean (SD) MADRS anhedonia factor score at induction phase baseline was 18.63 (3.02; $n = 603$); mean (SD) change from induction phase baseline to end of induction phase (day 28) was -11.81 (3.56; $n = 579$) and to end of O/M phase (week 48) was -13.11 (4.32; $n = 139$). The percentage of participants achieving $\geq 50\%$ reduction in MADRS anhedonia factor score at day 28 was 85.1% (493/579) and at week 48 was 89.9% (125/139). The mean (SD) PHQ-9 item 1 score at induction phase baseline was 2.39 (0.75; $n = 603$); mean (SD) change from induction phase baseline to day 28 was -1.38 (0.95; $n = 573$) and to week 47 was -1.75 (0.97; $n = 160$).

Conclusions: In these post-hoc analyses, esketamine+OAD treatment resulted in greater improvements in anhedonia symptoms based on MADRS anhedonia factor score and PHQ-9 item 1 compared with placebo+OAD in the short-term TRANSFORM-2 study. Anhedonia symptoms also decreased over the course of the long-term SUSTAIN-2 study with esketamine+OAD, further supporting it as a treatment option in patients with TRD including anhedonia symptoms.

Keywords: anhedonia, treatment-resistant Major Depressive Disorder, Esketamine nasal spray

Disclosure: Johnson and Johnson, Employee, Self, Johnson and Johnson, Stock / Equity - Privately Held Company, Self.

AMONG WOMEN WITH AND WITHOUT A HISTORY OF PERIPARTUM DEPRESSION

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Background: Peripartum depression (PPD) affects approximately 19% of women and may be influenced by sensitivity to hormonal fluctuations. Understanding the neural and molecular factors related to mood dysregulation during experimentally induced hormonal states may elucidate mechanisms underlying PPD. To interrogate the neural correlates of mood dysregulation in women at risk for PPD, this study performed an exploratory secondary analysis of a scaled-down model of peripartum hormone fluctuations, examining hormone-induced changes in brain functional connectivity (FC) and how those changes relate to molecular-genetic brain atlases.

Methods: Participants in this study (NCT04051320) included euthymic women with ($n = 11$) and without a history of PPD ($n = 12$), who received a baseline leuprolide acetate injection (Lupron; 3.75mg) to induce stable hypogonadism. Following one month in a hypogonadal state, participants began a two-week hormone “addback”, using micronized estradiol and progesterone tablets (4mg/day and 400mg/day, respectively). FC was assessed at baseline and at the conclusion of addback on a 7T Siemens MR scanner. Depression was defined as the magnitude of hormone-sensitive mood dysregulation, specifically, the peak increase in depression scores during addback or hormone withdrawal (i.e., hormone challenge) relative to follicular, pre-treatment baseline (Δ depression). The multivariate voxel pattern analysis (MVPA) -- and subsequent seed-based connectivity (SBC) analysis -- contrast identified regions where Δ FC- Δ depression associations differed by PPD history. Further, SBC maps were correlated with Allen Human Brain Atlas gene expression and PET receptor density atlases.

Results: MVPA identified a significant cluster in the posterior cingulate gyrus associated with increased depression during hormone challenge, which was moderated by PPD history. Post-hoc SBC revealed that, at baseline compared to hormone challenge, women with PPD and higher peak depressive symptoms exhibited greater negative connectivity between the posterior cingulate and bilateral dorsolateral prefrontal cortex, and increased connectivity from the posterior cingulate gyrus to the left supramarginal gyrus, right primary sensory cortex, and left premotor cortex (p 's < 0.001). Spatial correlation analyses demonstrated that unthresholded SBC maps were significantly associated with the expression of HSD3B1 ($r = -0.37$, $p = 0.01$) as well as 5-HT2a ($r = -0.47$, $p < 0.01$) and NET ($r = 0.485$, $p < 0.01$) receptor density.

Conclusions: In women with a history of PPD, dynamic changes in connectivity between the posterior cingulate gyrus and bilateral dorsolateral prefrontal cortices, along with changes involving frontoparietal sensorimotor regions, may contribute to mood dysregulation during peripartum hormone fluctuations. Moreover, the spatial expression of the HSD3B1 gene, 5-HT2A receptors, and NET receptors was significantly correlated with these changes in FC. HSD3B1 is an enzyme essential for hormone biosynthesis, specifically converting pregnenolone to progesterone, and this conversion has been linked to the development of PPD. Both 5-HT2A (serotonin 2A receptor) and NET (norepinephrine transporter) have shown altered binding in depression and are key targets of antidepressant medications. Taken together, these findings underscore the need to further investigate how neural and molecular alterations interact to confer susceptibility to PPD.

P224. NEUROBIOLOGICAL MARKERS OF HORMONE SENSITIVITY IN RESPONSE TO HORMONE MANIPULATION

Keywords: Maternal Depression, fMRI, gene expression, molecular mechanisms, hormone manipulation

Disclosure: Nothing to disclose.

P225. FEED FORWARD: APPETITE STRATIFIES ANTI-INFLAMMATORY TARGETING OF FATIGUE IN DEPRESSION

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Background: Fatigue is among the most common and disabling symptoms of depression, affecting 70–90% of patients across physical, cognitive, and social domains. It undermines quality of life, predicts poor functional outcomes, often leads to treatment resistance, and increases risk of relapse. Increasing evidence links fatigue to inflammation and metabolic dysregulation—an immunometabolic change that diverts energy away from behavioral priorities toward inflammatory demands, with changes in appetite reflecting the body's attempt to rebalance energy availability. TNF and related cytokines are central mediators of this axis, altering both behavioral regulation and metabolic pathways. Infliximab, a monoclonal antibody that blocks TNF- α , has shown preliminary benefits in treating treatment-resistant depression (TRD) with elevated inflammation; however, fatigue has not been systematically examined as an outcome in these studies, and no biomarker currently guides treatment selection. Objective: We tested whether baseline appetite change—a readily observable, mechanistically relevant marker of immunometabolic dysfunction—predicts fatigue response to infliximab versus placebo in TRD.

Methods: Sixty medically stable adults with TRD were randomized to infliximab ($n=30$) or placebo ($n=30$) across 12 weeks, with infusions administered at baseline, week 2, and week 6. Appetite status (increase vs decrease) was determined at baseline using the Inventory of Depressive Symptomatology–Self Report. Fatigue was assessed across nine study visits with the Multidimensional Fatigue Inventory (MFI), which captures five domains: physical, mental, general fatigue, reduced activity, and reduced motivation. Treatment $\times$ appetite $\times$ time (3-way) interactions were tested using mixed-effects models with piecewise trajectories (0–2, 2–6, and 6–12 weeks), while adjusting for age, sex, BMI, and race. An unstructured covariance matrix was selected based on Akaike's information criteria. Effect sizes were standardized to baseline standard deviations.

Results: At baseline, half of the patients ($n=31$) reported increases, and the remaining ($n=29$) reported decreases in appetite, with no differences in fatigue between groups ($p>0.64$). An overall effect of 3-way interactions was noted for total MFI scores (peak difference -18.2 points, $p=0.04$, Cohen's $d=1.17$), with infliximab benefiting subjects with increased appetite patients (peak $d=-0.86$) and placebo (peak $d=1.04$) benefiting subjects with decreased appetite. Physical fatigue demonstrated significant early effects ($p=0.03$), with infliximab superiority among patients with increased appetite at Weeks 2, 3, and 4 (all $p<0.05$; d ranging from -0.72 to -0.73). In contrast, depressed subjects with increased appetite demonstrated significant improvements in mental fatigue at mid-phase ($p=0.015$, $d=0.9$) and improvements in general fatigue with large effect sizes at mid-to-late phases ($p=0.018$, peak $d=1.36$). Across all dimensions, clinically meaningful effects ($|d|\geq 0.5$) were observed in 37 of 90 contrasts (41%), with patients consistently showing an infliximab benefit at 6–8 timepoints per fatigue dimension. Opposing effects favoring the placebo were shown in subjects with decreased appetite. Notably, reduced activity and motivation showed no significant interactions, suggesting appetite

specifically modulates energy-related rather than motivational fatigue dimensions.

Conclusions: Increased appetite emerges as a novel behavioral predictor of anti-inflammatory treatment response for fatigue in TRD. The temporal pattern revealed early physical improvements (Week 2) followed by mental and general fatigue benefits (Weeks 3–6), with sustained effects through Week 12, associated with large effect sizes. The dimension-specific and temporally ordered response pattern (physical, mental, general fatigue) may reveal an underlying neurobehavioral cascade of cytokine effects on immunometabolic pathways. The consistent pattern of infliximab benefit for increased appetite versus placebo benefit for decreased appetite suggests appetite changes reflect distinct immunometabolic phenotypes that modulate treatment efficacy. Appetite-based stratification may facilitate precision medicine for inflammation-related fatigue, establishing appetite assessment as a valuable biomarker for identifying patients likely to benefit from anti-inflammatory treatments.

Keywords: Fatigue, Appetite, Anti-inflammatory drugs, Immunometabolism, Energy

Disclosure: Nothing to disclose.

P226. SEX-SPECIFIC THALAMIC SUBREGIONAL DYSCONNECTIVITY IN FIRST-EPISODE DRUG-NAÏVE PATIENTS WITH MAJOR DEPRESSIVE DISORDER

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Background: The thalamus is a key subcortical structure implicated in the pathophysiology of major depressive disorder (MDD), with its functional connectivity (FC) linked to depressive symptoms and cognitive dysfunction. However, it remains unclear whether emotion and cognition of sex difference in MDD patients are associated with altered thalamocortical FC patterns.

Methods: We recruited 222 non-comorbidity, drug-naïve MDD patients and 159 matched healthy controls (HC). All participants were scanned by a 3.0T MRI scanner (Trio Tim, Siemens AG, Erlangen, Germany) with an 8-channel phased-array head coil. To explore changes in seed-based thalamus and thalamic subregional resting-state functional connectivity (rsFC) across all subjects, we used a fine-grained thalamic atlas with eight sub-regions per hemisphere including: the medial prefrontal thalamus (mPFTtha), the pre-motor thalamus (mPMtha), the sensory thalamus (Stha), the rostral temporal thalamus (rTtha), the posterior parietal thalamus (PPtha), the occipital thalamus (Otha), the caudal temporal thalamus (cTtha), and the lateral prefrontal thalamus (IPFtha). Thalamic subregions were defined using the Human Brainnetome Atlas, which provides a fine-grained cross-validated atlas and contains information on both anatomical and functional connections. We conducted a voxel-wise full-factorial analysis of variance analysis for bilateral whole and subregional thalamic FC separately, with diagnosis and sex both as the between-group factors, with age, education years, and mean FD as covariates to control their possible confounding effects. To determine group difference in different sex category on each thalamic FC pattern, the interaction effects between diagnosis (MDD vs HC) and sex (female vs male) were examined. The statistical threshold is $P<0.005$ at the voxel level, and family-wise error (FWE) corrected $P<0.05$ at the cluster level with cluster size >100 . Partial correlations assessed associations between altered FC and clinical or cognitive variables.

Results: Main effect of diagnosis on thalamic FC

Compared to HCs, patients with MDD showed increased FC between the right Stha and right superior temporal gyrus (STG) ($p < 0.001$, FWE-corrected, Figure 1B and Supplementary Table S1). The rsFC of the whole or other subregional thalamic FC did not differ significantly between groups.

Sex-by-diagnosis interactions on thalamic FC

Significant sex-by-diagnosis interactions were observed in the rsFC between the left mPMtha and right middle cingulate cortex (MCC) ($p = 0.001$, FWE-corrected), left IPFtha, and right MCC ($p = 0.037$, FWE-corrected), left Otha and bilateral supplementary motor area (SMA) ($p = 0.010$, FWE-corrected), left Otha and right middle frontal gyrus (MFG) ($p = 0.006$, FWE-corrected), and left Otha and right inferior temporal gyrus (ITG) ($p = 0.044$, FWE-corrected) (Table 2 and Figure 2A). No significant sex-by-diagnosis interactions were observed in the whole or other subregional thalamic FC between groups.

Post-hoc analysis revealed female patients with MDD showed significantly decreased FC between left mPMtha/IPFtha and right MCC, as well as between left Otha and bilateral SMA, left Otha and right ITG/MFG, compared to female HCs (P Bonferroni-corrected < 0.05 , Figure 2B). In contrast, male patients with MDD showed significantly increased FC in these same thalamocortical pathways relative to male HCs (P Bonferroni-corrected < 0.05 , Figure 2B).

Correlations between thalamic FC and clinical features

In male patients with MDD, we observe significant positive correlations between Stroop CN (s) and the rsFC of between left mPMtha and right MCC ($r = 0.347$, $p = 0.002$) and the rsFC of between left IPFtha- and right MCC ($r = 0.299$, $p = 0.007$), and a significant negative correlation between TMT-r scores and the rsFC of between the left Otha and right ITG ($r = -0.243$, $p = 0.031$). In female patients with MDD, we observe a significant negative correlation between psychic symptom scores and the rsFC of between the left Otha and right ITG ($r = -0.184$, $p = 0.027$) (Figure 3B). Further, the correlation between Stroop CN (s) and the rsFC of between left mPMtha and right MCC ($Z = 2.357$, $p = 0.018$), the rsFC of between left IPFtha and right MCC ($Z = 2.096$, $p = 0.036$), between TMT-r scores and the rsFC of between left Otha and right ITG ($Z = -2.435$, $p = 0.015$), and between psychic symptom scores and the rsFC of between left Otha and right ITG ($Z = 2.017$, $p = 0.044$) were significantly different between male and female with MDD.

Conclusions: These findings represent the first evidence of sex-specific thalamic subregional FC alterations in patients with MDD and these alterations were associated with symptoms and cognitive functions in a sex-specific manner. Specifically, reduced FC correlated with increased anxiety level in female subjects and increased thalamocortical FCs were associated with better cognitive performance in males. The identification of opposite FC alterations in male and female MDD underscored the need to consider sex as a key biological variable in both mechanistic research and treatment development.

Keywords: Major Depression Disorder, Thalamus, Resting State Functional Connectivity, thalamic subregional connectivity

Disclosure: Nothing to disclose.

P227. EFFICACY AND SAFETY OF INHALED MEBUFOTENIN (GH001) IN PATIENTS WITH BIPOLAR II DISORDER AND A CURRENT MAJOR DEPRESSIVE EPISODE: RESULTS FROM A PHASE 2A CLINICAL TRIAL

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Background: Bipolar II disorder (BDII) is a chronic psychiatric disorder marked by recurring episodes of hypomania and major depressive episodes (MDE), that places a significant burden on affected individuals. The lifetime prevalence of BDII is estimated to range between 0.4 and 5%. Current treatments for depressive symptoms in patients with BDII remain limited, offering insufficient efficacy and tolerability, highlighting the need for new therapeutic approaches. Mebufotenin (5-methoxy-N,N-dimethyl-tryptamine [5-MeO-DMT]) is a rapid acting psychoactive molecule that acts as a non-selective serotonin agonist with highest affinity for the 5-HT_{1A} receptor subtype. GH001, a synthetic form of mebufotenin for pulmonary inhalation, has been well tolerated in early-stage trials in healthy volunteers and patients with TRD, with a rapid reduction in the severity of depressive episodes. The present trial is the first to evaluate mebufotenin in patients with BDII experiencing a current MDE, focusing on the safety and antidepressant potential of GH001 in this population.

Methods: This Phase 2a, proof-of-concept, open-label trial enrolled patients aged 18–64 years (inclusive) who met the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5) diagnostic criteria for BDII and a current MDE, and who had a Montgomery-Åsberg Depression Rating Scale (MADRS) score of ≥ 24 at pre-dose (NCT05839509). Patients were not permitted to receive any antidepressant medications (including selective serotonin reuptake inhibitor, serotonin and norepinephrine reuptake inhibitor, and tricyclic antidepressant) within 7 days or 5 half-lives, whichever was longer, prior to dosing. Lithium use within 6 months prior to dosing was not permitted, if applicable. GH001 was administered via inhalation as an individualized dosing regimen (IDR) of at least one and up to three escalating doses (6, 12, and 18 mg) on a single day (Day 1). The criteria for administration of the second and third doses were based on the patient's subjectively reported psychoactive effects (PsE), and the safety and tolerability at the previous dose level according to the trial physician's judgement. This trial was conducted under the supervision of qualified healthcare professionals, providing psychological support per standard-of-care, but without any planned psychotherapeutic intervention before, during, or after dosing. The primary endpoint was the change in MADRS from baseline to Day 8. Other efficacy endpoints assessed included response ($\geq 50\%$ reduction from baseline in MADRS total score), remission (MADRS total score ≤ 10), Clinical Global Impression-Severity (CGI-S), and Bipolar Depression Rating Scale (BDRS). The safety and tolerability of GH001 were evaluated up to Day 8 in addition to manic symptoms assessed by the Young Mania Rating Scale.

Results: A total of six patients aged 32–57 with BDII and a current MDE were enrolled in this trial. Mean (SD) MADRS total score at baseline was 32.0 (5.1). The primary endpoint was achieved, with a significant reduction from baseline to Day 8 with a mean (SD) MADRS total score of -16.8 (11.1) which corresponds to a reduction of 52.5% ($P = 0.0099$). Significant reductions in the mean (SD) MADRS total score were also observed at 2-h post-dose (-16.3 [6.0; $P < 0.0006$]) and on Day 2 (-13.3 [13.5; $P < 0.0299$]). One-third (33.3%) of patients showed response to treatment on Day 8, with a remission rate of 33.3% at Day 8. The rapid reduction in depressive symptoms as assessed by the MADRS was mirrored in the CGI-S and the BDRS, with a mean (SD) reduction of -2.5 (1.5) on the CGI-S and -14.5 (11.2) on the BDRS observed from baseline and Day 8. Inhalation of GH001 was well tolerated and no treatment-related serious adverse events were reported. The majority of treatment-emergent adverse events were mild (83.3%) or moderate (11.1%) in severity, with the most commonly reported events being headache (50.0%), nausea (33.3%), and anxiety (33.3%), and all other events were only reported once. Following dosing with GH001, YMRS scores remained low and stable, decreasing

from 2.2 at baseline to 1.0 by Day 8 (-1.2 [$SD = 1.5$]), indicating no emergence of manic symptoms.

Conclusions: In this trial, GH001 induced rapid and significant improvement in the symptoms of depression in patients with BDII and a current MDE and was well tolerated.

Keywords: Bipolar II disorder, mebufotenin, psychoactive molecules

Disclosure: AbbVie, Honoraria, Self, Boehringer Ingelheim, Honoraria, Self, Cycleron, Honoraria, Self, Compass, Honoraria, Self, GH Research, Honoraria, Self, Janssen, Honoraria, Self, LivaNova, Honoraria, Self, Medice, Honoraria, Self, MSD, Honoraria, Self, Newron, Honoraria, Self, Sage/Biogen, Honoraria, Self, Shire/Takeda, Honoraria, Self, Medice, Grant, Self, Janssen, Grant, Self.

P228. RAPID ANTIDEPRESSANT EFFECTS OF INHALED GH001 IN TREATMENT-RESISTANT DEPRESSION: RESULTS FROM A PHASE 2B, DOUBLE-BLIND, RANDOMIZED CONTROLLED TRIAL WITH 6-MONTH FOLLOW-UP

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Background: Treatment-resistant depression (TRD) represents a particularly challenging form of major depressive disorder (MDD), defined by an inadequate response to at least two antidepressants given at adequate dose and duration within the current depressive episode. TRD affects nearly one-third of individuals with MDD and is consistently linked to greater psychiatric and medical comorbidity, increased rates of hospitalization, elevated suicide risk, and a significantly reduced quality of life compared to non-resistant depression. There are currently only two pharmacotherapies approved for TRD in the United States (esketamine and olanzapine-fluoxetine combination) highlighting the significant unmet need for fast-acting, effective, and safe treatments. Mebufotenin (5-methoxy-N,N-dimethyltryptamine [5-MeO-DMT]) is a highly potent naturally occurring psychoactive substance from the tryptamine class which acts as a non-selective serotonin (5-HT) agonist with the highest affinity for the 5-HT_{1A} receptor subtype. Evidence from early-phase trials suggest that GH001, a synthetic form of mebufotenin for pulmonary inhalation, is well tolerated in healthy volunteers and patients with TRD and may induce rapid improvements in depressive symptoms.

Methods: This Phase 2b multicenter trial assessed the efficacy and safety of GH001 in patients with TRD (NCT05800860). Part 1 was a 7-day, randomized, double-blind, placebo-controlled part where patients were randomized in a 1:1 ratio to receive GH001 or placebo. Part 2 was a 6-month open-label extension (OLE) where up to five GH001 re-treatments were administered, based on the patients Montgomery-Åsberg Depression Rating Scale (MADRS) scores, and the safety and tolerability of the previous dose(s). On Day 8 of Part 1, all patients directly transitioned to Part 2. In both parts, GH001 (or placebo in Part 1) was administered as an individualized dosing regimen (IDR) of up to three escalating doses of GH001 (6, 12 and 18 mg, or placebo) administered on a single dosing day with a 1-hour interval between doses. The criteria for administration of the second and third doses in the IDR were based on the patient's subjectively reported psychoactive effects (PsE), and the safety and tolerability at the previous dose level, according to the trial physician's judgement. This trial was conducted under the

supervision of qualified healthcare professionals, providing psychological support per standard-of-care, but without any planned psychotherapeutic intervention before, during, or after dosing. Patients attended regular scheduled visits during the OLE. The primary endpoint of this trial was the mean change in MADRS from baseline to Day 8, assessed by an independent blinded rater. Patients aged 18–64 with a Hamilton Depression Rating Scale total score ≥ 20 at screening and baseline, and nonresponse ($\leq 25\%$ improvement) to ≥ 2 and ≤ 5 oral antidepressant treatments started during the current major depressive episode, were eligible to enroll in this trial.

Results: A total of 81 patients with TRD were enrolled in Part 1 with 40 patients randomized to GH001 and 41 randomized to placebo. Change in MADRS total score from baseline to Day 8 was significantly greater with GH001 versus placebo (least squares mean difference [standard error], -15.5 [1.7]; $P < 0.0001$) with an effect size (Cohen's d) of -2.0 . Statistically significant reductions were also observed in the GH001 group at 2 h post-dose and on Day 2. On Day 8, remission (MADRS total score ≤ 10) was achieved in 57.5% of patients in the GH001 group versus 0% in the placebo group ($P < 0.0001$). Of the 63 patients who completed Part 2, 73.0% ($n = 46$) were in remission at 6 months and this was achieved with a mean of four treatments, with 63.5% ($n = 40$) of patients requiring 1–4 treatments in 6 months. The median duration of PsE across all doses and treatment visits was 11 minutes. Inhalation of GH001 was well tolerated, and most adverse events (AEs) were mild or moderate with no treatment-related serious AEs (SAEs). One treatment-emergent SAE (preferred term migraine) occurred during the OLE and was deemed unrelated to treatment. Over the 6-month duration of the trial, lower rates of suicidal ideation compared to baseline were observed at each timepoint assessed with no treatment-emergent events of suicidal intent or suicidal behavior. At 99.0% (285/288) of treatment visits, patients were considered to be discharge ready at 1-h post-dose on the dosing day.

Conclusions: GH001 treatment was well tolerated and demonstrated rapid and significant improvements in depressive symptoms in patients with TRD. Data from Part 2 suggest that GH001 can maintain long-term remission in TRD, with 73.0% of patients who completed the OLE in remission at 6 months with infrequent retreatments. The brevity of the PsE, along with rapid symptomatic improvement, and favorable safety profile may align with patient and provider preferences and permit scalable clinical use. These results support the evaluation of GH001 in larger populations in Phase 3 trials.

Keywords: Treatment-resistant depression, mebufotenin, psychoactive molecules

Disclosure: Johnson and Johnson, Advisory Board, Self, Abbvie, Advisory Board, Self, Otsuka, Advisory Board, Self, Johnson and Johnson, Speakers Bureau, Self, GH, Advisory Board, Self, GH, Consultant, Spouse/Partner, Johnson and Johnson, Consultant, Spouse/Partner.

P229. MORE THAN A MISSED REWARD: NEURAL SIGNATURES OF REWARD PROCESSING ACROSS APPETITIVE PROFILES IN MAJOR DEPRESSIVE DISORDER

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Background: Reward processing deficits are prominent in major depressive disorder (MDD) and contribute to appetitive subtypes: hyperphagic (HyperMDD) and hypophagic (HypoMDD). However, few studies examine neurobiological processes underlying these subtypes, and whether they stem from aberrant responsivity specific to food or more broadly across different rewards. The current study examined group differences in functional connectivity during the anticipation and outcome phases of food vs. monetary rewards across MDD appetitive phenotypes. Hypotheses: 1) Relative to healthy controls (HCs), the HyperMDD group would show altered mesocorticolimbic reward system functional connectivity when anticipating food (vs. monetary) rewards, while the HypoMDD group would exhibit reduced connectivity during anticipation of both reward types. 2) The HyperMDD group would demonstrate altered mesocorticolimbic reward system connectivity when processing food (vs. monetary) reward outcomes, with connectivity patterns being most pronounced during failed reward outcomes.

Methods: 14 unmedicated HyperMDDs, 20 unmedicated HypoMDDs, and 36 HCs, matched on age, sex and BMI, completed food and monetary incentive delay tasks (FID, MID) while undergoing fMRI, ~1.5 h after a standardized meal. Psychophysiological interaction (PPI) analyses in CONN (22.v2407) examined functional connectivity changes during relevant task conditions according to reward processing components: anticipation (Food Cue, Money Cue), successful reward receipt (Successful Food Outcome, Successful Money Outcome), and failed reward receipt (Failed Food Outcome, Failed Money Outcome). Second-level seed-to-voxel analyses examined interactions between group and reward type. ROIs, including the nucleus accumbens (NAcc), orbitofrontal cortex (OFC), ventromedial prefrontal cortex (vmPFC), anterior insula (aINS), thalamus, and dorsal anterior cingulate cortex (dACC), were anatomically defined using the Automated Anatomical Labeling 3 atlas. Significant effects were defined as $p(\text{FDR}) < 0.05$. ANOVAs tested group differences in behavioral and clinical data and exploratory Pearson correlations tested associations between caloric intake during a post-FID snack consumption period, BDI scores and functional connectivity patterns during anticipatory food reward processing.

Results: HyperMDDs responded faster to food reward cues than HCs ($p = 0.044$). Groups did not differ in caloric intake during the post-FID snack consumption period ($p = 0.733$). PPI analyses revealed group differences in connectivity during anticipatory reward processing according to reward type (Food Cue vs. Money Cue); the HyperMDD group demonstrated negative OFC-precentral gyrus connectivity (decoupling) relative to HCs (Hyper > HC: $t(1,67) = -5.90$, $p(\text{FDR}) = 0.000000$) for Food Cues and increased connectivity for Money Cues, while HCs showed similar OFC connectivity for Money and Food Cues. For the aINS seed, the HypoMDD group displayed positive aINS-DLPFC connectivity for Food Cues and negative coupling for Money Cues, while the reverse was observed for HCs (Hypo > HC: $t(1,67) = 5.63$, $p(\text{FDR}) = 0.000001$). Finally, the HypoMDD group exhibited increased dACC-thalamus/hippocampus connectivity for Food Cues together with decoupling for Money Cues (Hypo > HC: $t(1,67) = 5.04$, $p(\text{FDR}) = 0.000011$), while the HyperMDD group showed dACC-thalamus/hippocampal decoupling for Food Cues relative to Money Cues (Hyper > Hypo: $t(1,67) = -5.45$, $p(\text{FDR}) = 0.000001$). Group differences in connectivity were also observed during the receipt phase (Successful Food Outcome vs. Successful Money Outcome). Compared to HCs, the HypoMDD group demonstrated positive coupling between NAcc-temporal pole (Hypo > HC: $t(1,67) = 5.29$, $p(\text{FDR}) = 0.000003$), NAcc-postcentral gyrus (Hypo > HC: $t(1,67) = 4.34$, $p(\text{FDR}) = 0.000050$) and thalamus-middle temporal gyrus (Hypo > HC: $t(1,67) = 4.86$, $p(\text{FDR}) = 0.000007$) for successful food receipt, and decoupling for successful money receipt. The HyperMDD group, relative to HCs, demonstrated aINS-medial frontal cortex decoupling during successful food receipt

(Hyper > HC: $t(1,67) = -5.29$, $p(\text{FDR}) = 0.000001$), and positive coupling during successful money reward receipt. For failed receipt outcomes (Failed Food Outcome vs. Failed Money Outcome), the HyperMDD group demonstrated NAcc-middle frontal gyrus (Hyper > Hypo: $t(1,67) = -5.24$, $p(\text{FDR}) = 0.000003$), dACC-lateral occipital cortex (Hyper > HC: $t(1,67) = -4.85$, $p(\text{FDR}) = 0.000008$), and thalamus-parietal operculum (Hyper > HC: $t(1,67) = -5.58$, $p(\text{FDR}) = 0.000001$) decoupling for Failed Food Outcomes, and positive coupling for Failed Money Outcomes. Caloric intake and BDI scores were not associated with connectivity in the full sample or between MDD groups.

Conclusions: Findings reveal dissociable patterns of mesocorticolimbic connectivity during food vs. monetary reward processing in MDD appetitive subtypes. HypoMDD was associated with heightened connectivity to food and negative connectivity to money, while the HyperMDD group exhibited the opposite: reduced coupling to food reward cues and positive coupling to monetary rewards. These findings suggest that appetitive MDD subtypes are distinguished not by global reward dysfunction, but by opposing circuitry profiles that bias responsivity toward food versus monetary rewards, patterns that may help guide the development of subtype- and reward-specific interventions.

Keywords: Depression subtypes, Anticipatory reward activation, Monetary reward, food reward, monetary incentive delay task

Disclosure: Nothing to disclose.

P230. EXPLAINABLE AI-DERIVED PATIENT PERSONAS REVEAL NEUROBIOLOGICAL PREDICTORS OF KETAMINE RESPONSE IN TREATMENT-RESISTANT DEPRESSION

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Background: Ketamine has emerged as a promising therapy for treatment-resistant depression (TRD), yet its acute psychoactive effects complicate blinding and make it difficult to disentangle true antidepressant response from expectancy. Identifying reproducible neurobiological signatures of ketamine responders may clarify its effectiveness for treating depression, increase trial interpretability, and enable development of precision treatment strategies.

Methods: We reanalyzed data from a small, double-blind, placebo-controlled, crossover Phase II trial of intravenous racemic ketamine (0.5 mg/kg) versus saline placebo (NCT0008869; $n = 33$). NetRAI, an explainable machine learning (ML) framework, was applied to this dataset. With this approach the patient population is broken into explainable (ketamine-response and placebo-response) sub-groups and unexplainable subgroups to identify high-effect-size subpopulations (Personas) characterized by 2–4 baseline or screening variables.

Due to the limited sample size and the crossover trial design, participant data from both infusion sessions were pooled, yielding 63 analyzable cases (3 incomplete data entries were excluded). Treatment response was defined as a $\geq 40\%$ reduction in MADRS scores at Day 7. Variables included clinical outcomes (MADRS, HAMA, HAM-D-17, BDI), physiological measures (e.g., BMI), volumetric MRI features, and resting-state MEG recordings. Personas were identified in a 50/50 train ($n = 32$)/test ($n = 31$), validated with bootstrapping, and meeting significance thresholds ($p < 0.05$; Cohen's $d > 0.5$).

Results: Several Personas emerged that were identified in the training set and reproduced in the testing set that defined distinct

ketamine responder subgroups using clinical scale and MRI variables:

Persona 1: White matter-emotional regulation ($p = 0.0286$, Cohen's $d = 2.1153$).

Persona 1 from Training set ($n = 14$; Placebo = 7 [CI 1.14–4.86], Ketamine = 7 [CI 8.71–20.00])

Replicated Persona 1 in the Testing set ($n = 8$; Placebo = 4 [CI (–3.00)–(7.50)], Ketamine = 4 [CI 12.50–24.50])

Characterized by lower left posterior cingulate white matter volume (3375.2–4522.7 mm³; data range: 3375.2–6573.9 mm³), lower right inferior parietal white matter volume (8199.2–12663.9 mm³; data range: 8199.2–15082.6 mm³), and low emotional reactivity on the BDI Crying item (0–1; data range 0–3). Patients with this profile showed preferential benefit from ketamine, consistent with findings regarding ketamine and default mode network emotional regulation circuits and emotional reactivity.

Persona 2: Grey matter-interoceptive/autonomic ($p = 0.0476$, Cohen's $D = 1.3309$).

Persona 2 from Training set ($n = 13$; Placebo = 7 [CI (–2.85)–2.14], Ketamine = 6 [CI 4.33–16.83])

Replicated Persona 2 in the Testing set ($n = 12$; Placebo = 6 [CI (–3.33)–(–0.17)], Ketamine = 6 [CI 0.17–19.17])

Characterized by higher left isthmus cingulate grey matter volume (2505–3742 mm³; data range: 1824–3742 mm³), higher right lingual grey matter volume (6805–9041 mm³; data range: 4454–9041 mm³), and low cardiovascular anxiety symptoms on HAMA (0–1; data range 0–3). This phenotype implicates ketamine's modulation of salience and interoceptive networks, supporting a mechanism of autonomic recalibration.

Additional Personas were identified using MEG-derived network metrics, highlighting connectivity changes to further support circuit-level mechanisms of responses

Conclusions: By integrating clinical, physiological, structural, and functional neurobiology, we identified reproducible patient Personas that map onto ketamine's mechanistic effects in TRD. These phenotypes suggest ketamine exerts antidepressant activity through at least two neurobiologically distinct pathways: (1) restoration of emotional regulation circuits via posterior cingulate-parietal white matter networks and (2) recalibration of interoceptive/autonomic processing via cingulate-lingual grey matter and salience circuitry. These findings support precision psychiatry approaches for patient stratification and clinical trial enrichment in TRD.

Key Findings:

Keywords: major depression, machine learning, (R,S)-ketamine, CNS Clinical Trials, patient selection

Disclosure: Johnson and Johnson, Consultant, Self, Otsuka, Consultant, Self, Merck, Consultant, Self, NetraMark, Employee, Self, Neumarker, Consultant, Self.

P231. A PHASE IB STUDY TO EVALUATE THE PHARMACODYNAMICS, SAFETY, AND TOLERABILITY OF THE NOVEL NEUROPLASTOGEN ZALUPINDOLE (DLX-001), DOSED DAILY AND INTERMITTENTLY IN PARTICIPANTS WITH MAJOR DEPRESSIVE DISORDER

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Background: Zalsupindole is a non-hallucinogenic, non-dissociative neuroplastogen being developed for the treatment of major depressive disorder (MDD). Here, we describe findings from a phase Ib dose-blinded, single-center, multiple-dose study in participants with recurrent MDD. This study was designed to

assess the effects of zalsupindole on translational biomarkers associated with neuroplasticity, as well as safety, tolerability, and preliminary efficacy. Participants in Cohort A ($n = 9$) received zalsupindole once daily for 7 days, and participants in Cohort B ($n = 9$) received zalsupindole twice over 7 days.

Methods: Upon admission to a research clinic (Groningen, NL), participants completed habituation and baseline assessments, including polysomnography (PSG), quantitative electroencephalography (qEEG), and clinical symptom scales. In Cohort A, participants received individualized CYP2D6-adjusted doses of zalsupindole once daily for 7 days. On Days 1, 4, and 7, qEEG was collected while awake; on the evenings of Day 1 and Day 7, PSG was collected overnight. In Cohort B, participants received zalsupindole once on Days 1 and 4. After discharge from the unit on Day 8, participants returned for assessments on Days 22 and 36.

Results: Mean age at admission was 33 years (range 23–54) for Cohort A and 32 years (range 20–46) for Cohort B, with 5/9 (55%) females in each cohort. Mean HAM-D of 18.8 (SD = 2.5) for Cohort A and 17.8 (SD = 2.3) for Cohort B indicated moderate depression severity at baseline. Co-morbid anxiety measured by the HAM-A was low at baseline (Cohort A: Mean = 12.0, SD = 4.8; Cohort B: Mean = 13.0, SD = 6.0). All 18 participants completed the treatment period. Zalsupindole was well-tolerated. The most frequent zalsupindole-related AEs were self-limited nausea ($n = 9$) and headache ($n = 9$). There were no changes in safety parameters, including vital signs, ECGs, laboratory measures, or physical exam findings. Consistent with the previous phase I healthy participant study, there was no evidence of psychotomimetic, hallucinatory, or dissociative effects, as measured by the MEQ-30, BPRS, and CADSS. Plasma levels of zalsupindole were consistent with target exposures. qEEG from both cohorts demonstrated increases in slow-wave delta and theta activity. From an efficacy perspective, rapid, meaningful, and sustained decreases in depression scores were observed in both cohorts. Mean change from baseline on the MADRS was 10.8 points (SD = 8.7) at Day 8 and 11.5 points (SD = 5.9) at Day 36 in Cohort A, and 12.6 points (SD = 6.7) at Day 8 and 9.6 points (SD = 6.9) at Day 36 in Cohort B, demonstrating that improvement on the MADRS was both rapid (measurable at the first post-treatment assessment) and sustained for 4+ weeks after the last dose of zalsupindole was administered.

Conclusions: This PhIb study's primary endpoint—change from baseline to end of treatment on qEEG slow-wave activity—is an emerging translational marker of cortical plasticity and synaptic strength (Duncan et al. 2013). Data from both cohorts demonstrated a consistent increase in slow-wave delta and theta activity, providing further evidence that promotion of neuroplasticity may play a critical role in the efficacy of zalsupindole. Moreover, MADRS data support that zalsupindole—whether administered daily or intermittently—leads to rapid, robust, and sustained improvement in depressive symptoms in patients with recurrent MDD. The optimal dosing schedule for zalsupindole remains to be determined, with both daily and intermittent dosing schemes planned to be evaluated in a forthcoming Ph2 study.

Keywords: neuroplastogen, Major Depressive Disorder (MDD), Psychoplastogens, Human Clinical trial

Disclosure: Delix Therapeutics, Employee, Self, Merida Biosciences, Employee, Spouse/Partner.

P232. BRAIN NETWORK SIGNATURES OF DEPRESSION AND TREATMENT-RESISTANT DEPRESSION STUDIED WITH MAGNETOENCEPHALOGRAPHY

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Background: Depression is a serious mental health disorder affecting more than 21 million adults in the United States, with nearly 30% of those not responding adequately to standard pharmacological treatments, a condition known as treatment-resistant depression (TRD). To address limitations related to self-report and other biases, objective neurobiological markers for assessing major depressive disorder (MDD) and TRD are needed. Recent evidence suggests MDD is characterized by altered connectivity in large-scale brain networks including the default mode (DMN), executive control (ECN), and salience (SN) networks. Much of this work has been derived from modeling the brain's hemodynamic response—a measure of neural metabolic demand lacking temporal specificity. In contrast, magnetoencephalography (MEG) offers high-resolution mapping of neural electrical currents, and parcel-based orthogonalized envelope correlations can be used in tandem with MEG to measure how the amplitude of neural signals from different brain regions correlate—referred to as coherence. Studying coherence within canonical networks could offer promise for improving diagnostic precision in MDD and TRD and inform the development of targeted treatments.

Methods: 168 participants (88 DSM-IV or 5 MDD, 80 healthy volunteers, HV, mean age = 37.2 ± 12.5 years) were included in the study. Participants with MDD were further categorized into TRD (more than 2 treatment failures, $n = 74$) and those who were not treatment-resistant ($n = 14$). One or two eyes-closed resting-state scans were collected per participant using a CTF 275-channel MEG system. T1-weighted structural magnetic resonance imaging scans were collected and co-registered to the MEG data using fiducial landmarks. Source reconstruction was performed using a linearly constrained minimum variance beamformer in the theta (4–8 Hz), alpha (9–14 Hz), beta (15–29 Hz), and gamma (30–58 Hz) frequencies. Data were segmented into 200 parcels, orthogonalized, and Hilbert transformed to estimate pairwise envelope correlation coefficients in each bandwidth. Brain connectivity coherence was calculated between parcels within the default mode (DMN), executive control (ECN), and salience (SN) networks. Data Integration Analysis for Biomarker discovery using Latent variable approaches for Omics studies (DIABLO) was used to identify neural connectivities predictive of 1) MDD versus HV, and 2) TRD-only versus HV. A series of MANOVA analyses were conducted with group membership entered as an independent variable and connectivity scores entered as dependent variables, with biological sex, age, and ethnicity entered as covariates.

Results: The overall correlation structure between brain networks was estimated using both the discrimination and sparse partial least squares (sPLS) approaches. The discrimination model (area under the curve, AUC range 0.758–0.787) was found to produce higher prediction accuracy scores compared to the sPLS model (AUC range 0.611–0.646) and used for subsequent analyses. Across frequencies, the ECN most effectively distinguished MDD from HV in the theta and beta bands ($ps < 0.05$), while in the TRD-only model, the DMN (AUC = 0.849) outperformed the ECN (AUC = 0.707) but not the SN (AUC = 0.773) in the beta band. Within networks, we observed hyperconnectivity in patients with depression. In the MDD model, patients showed increased coherence between the orbitofrontal cortex and the precuneus ($p < 0.01$), temporal cortex ($p < 0.03$), and medial prefrontal cortex ($p < 0.04$) within the ECN in the beta band relative to the HV group. In the TRD-only model, patients showed increased coherence within the SN in the alpha band (between the superior parietal lobule and frontal operculum, $p < 0.04$) and within the SN in the gamma band (between the temporal cortex and insula, $p < 0.04$) compared to the HV group. In the beta band, the TRD group also exhibited increased coherence in the DMN (between the inferior parietal lobule and the precuneus, lateral prefrontal cortex, dorsal prefrontal cortex, and between the temporal pole and parahippocampal cortex, $ps < 0.02$), in the ECN (between the posterior cingulate cortex and frontal pole, as well as the temporal

cortex, and between the intraparietal sulcus and lateral prefrontal cortex, $ps < 0.04$), and in the SN (between the insula and lateral prefrontal cortex, and between the inferior frontal gyrus and frontal medial cortex, $ps < 0.05$) compared to the HV group.

Conclusions: Functionally, large-scale brain networks share overlapping roles in integrating cognitive, emotional, and sensory information, but the specific patterns of hyperconnectivity may reflect differences in underlying pathophysiology between depression and TRD. In depression, hyper-connectivity may represent compensatory mechanisms or maladaptive integration within core networks involved in self-referential processing and executive functioning, potentially worsening deficits in decision-making and emotion regulation. Our results show that canonical networks, including the DMN, ECN, and SN, can distinguish MDDs or TRDs from HVs, with each network contributing uniquely across frequency bands. These findings advance our understanding of the neural mechanisms underlying depression and treatment resistance, underscoring the value of innovative analytic approaches for future research and clinical applications.

Keywords: magnetoencephalography, Treatment-resistant depression, functional brain connectivity, computational psychiatry

Disclosure: Nothing to disclose.

P233. REAL-WORLD EVIDENCE DEMONSTRATES THE EFFECTIVENESS OF SAINT IN A HETEROGENOUS POPULATION: A MULTISITE, OPEN-LABEL STUDY

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Background: SAINT is a functional connectivity-guided, accelerated form of transcranial magnetic stimulation (TMS) that utilizes intermittent theta-burst stimulation (iTBS). SAINT is FDA-cleared for the treatment of major depressive disorder (MDD) and has reported high rates of remission in open label and randomized controlled trials. To evaluate the generalizability of SAINT in a real-world setting, an open-label, multi-center trial was conducted.

Methods: This study is part of an ongoing, multicenter clinical trial evaluating the real-world effectiveness of SAINT for the treatment of MDD. Participants (male and female) were enrolled across seven geographically diverse, community-based psychiatric clinics in the United States. This trial was pre-registered in the U.S. Clinical Trials registry (NCT05819021). Participants with a primary diagnosis of MDD, in a major depressive episode and who had failed to receive satisfactory improvement from a prior antidepressant medication in the current episode were enrolled. All participants received a 5-day course of SAINT. The primary outcome was the Clinical Global Impressions Scale of Improvement (CGI-I) score following the final treatment. Secondary outcome measures included the CGI-S to measure symptom severity rated by study site personnel and the Montgomery Asberg Depression Rating Scale (MADRS) rated immediately post-treatment by a third-party assessor.

This per protocol analysis was conducted following the closure of enrollment and after all participants completed the 5 days post-treatment endpoint. Clinically meaningful benefit was defined as a reduction in the CGI-S by 1 or more points. For results categorizing by treatment severity at baseline using the CGI-S, severity was categorized as: normal: 1, mild: 2–3, moderate: 4–5, severe: 6–7. Frequencies were compared using a Pearson's Chi-square test. Covariates examined for association with remission included age, sex, gender, study site, and years in current depressive episode,

and baseline CGI-S. Covariates were assessed with independent generalized linear models (GLMs) within the per-protocol population using the CGI-I remission status as a binary outcome. For post-treatment CGI-S differences, baseline values were compared to values collected within one week of completing treatment (the lower value of either the end of treatment day 5- or five-days post-treatment). For post-treatment MADRS differences, baseline values were compared to the five days post-treatment values. For CGI-S and MADRS differences, a two-sided, paired t-test with an alpha level of 0.05 was used.

Results: At this interim analysis milestone, 101 participants (56 male, 45 female) were enrolled and completed the 5-day course of SAINT per protocol. Participants had a mean of 19.0 (SD = 14.2) years since receiving a depression diagnosis and 3.5 (SD = 5.2) years in the current depressive episode.

Following treatment, 89.1% of participants achieved a clinically meaningful benefit (defined as improvement of ≥ 1 point on the CGI-S). Across the entire per protocol cohort, the primary outcome, mean CGI-I at the end of treatment day 5, was 2.32 with 68.3% of participants meeting the remission criteria (CGI-I ≤ 2) within one-week post-treatment. In categorizing participants by baseline symptom severity, remission criteria were met by 83.3% of participants with mild severity ($n = 6$), 70.9% with moderate severity ($n = 55$), and 62.5% with severe symptoms ($n = 40$). No statistical difference in remission rates between baseline severities was observed ($\chi^2(2) = 1.42$, $p = 0.49$). Remission rates also did not statistically differ between study sites ($\chi^2(6) = 5.70$, $p = 0.46$), and none of the assessed covariates remained significant after multiple comparisons when evaluated for their impact on remission status.

Consistent reductions in the CGI-S and MADRS were also observed following treatment. The CGI-S within one week of treatment was reduced compared to baseline (mean baseline/post-treatment difference = 2.21, SD = 1.47, 95% CI [1.92, 2.50], $t(100) = 15.146$, $p = 2.2e-16$, Cohen's $d = 1.53$, $n = 101$). Likewise, the post-treatment MADRS was reduced compared to baseline (mean baseline/post-treatment difference = 9.13, SD = 9.20, 95% CI [7.17, 11.03], $t(89) = 9.383$, $p = 6.048e-15$, Cohen's $d = 0.99$, $n = 90$). Taken together, these results demonstrate a consistent reduction in symptom severity as assessed by site clinicians (CGI-S) and third-party raters (MADRS).

Conclusions: This study was designed to evaluate SAINT in a naturalistic clinical setting, with inclusion and exclusion criteria representative of real-world use of TMS treatment. The SAINT protocol incorporates several adaptations beyond conventional TMS/iTBS, including fMRI-guided targeting and 10-h treatment days. This study demonstrates that community-based clinics can feasibly implement SAINT with high protocol fidelity (94% of participants treated per protocol). Clinical outcomes were consistent across sites, further supporting the generalizability and scalability of this approach for applications beyond academic environments. Results from this trial supports previously reported remission rates and clinically meaningful symptom reduction from the open label trial and RCT while extending these findings to more diverse and clinically representative populations.

Keywords: Transcranial Magnetic Stimulation (TMS), Accelerated intermittent theta burst stimulation (iTBS), Major Depressive Disorder (MDD), SAINT, fMRI-guided Targeting

Disclosure: Magnus Medical Inc, Founder, Self.

P234. SEROTONIN AND NEUROTICISM INTERACT TO MODULATE GUILT AND SHAME

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Background: Excessive guilt and shame can cause distress and impairment, often noticeable in depression and social anxiety. Recent work has shown that temperament traits can interact with serotonin function to modulate these social emotions. Neuroticism is a temperament trait that is widely regarded as a risk factor for depression and anxiety. People high in neuroticism have an emotional predisposition to expect negative outcomes. Since serotonin is involved in a wide range of psychological functions, including social emotions, here, we predicted that serotonergic state and neuroticism would interact to modulate feelings of guilt and shame.

Methods: We tested the effects of the dietary manipulation acute tryptophan depletion (ATD), which temporarily reduces brain serotonin synthesis by depleting its precursor, tryptophan, in a double-blind randomized placebo-controlled between-groups study of healthy volunteers (placebo $n = 36$, 17 female; ATD $n = 37$, 17 female). Guilt and shame were assessed via a computerized task that required mentally simulating social situations involving unjust harm. The participant was the agent of harm in half of the trials, and the victim of harm in the other half of trials. Neuroticism was measured via the NEU scale of the Structure of Temperament Questionnaire (STQ-77) and treated as a continuous variable throughout.

Results: Results for guilt ratings revealed a significant three-way interaction between serotonin status x neuroticism x agency ($F(1,69) = 11.146$, $p = 0.001$, $\eta^2 = 0.139$). Whereas under placebo, the guilt experienced when inflicting harm on others (as agent) increased with neuroticism ($r(36) = 0.509$, $p = 0.002$), the feeling of guilt was significantly alleviated by serotonin depletion. At the same time, in the victim's role, those higher in neuroticism felt more guilt following depletion than under placebo. For shame ratings, there was a significant three-way interaction between serotonin status x neuroticism x agency ($F(1,69) = 6.730$, $p = 0.012$, $\eta^2 = 0.089$). Follow-up analyses demonstrated that this was driven by individuals higher in neuroticism recording significantly higher shame ratings in situations where they were the victim after receiving depletion ($r(37) = 0.350$, $p = 0.034$).

Conclusions: Neuroticism interacted with serotonin function to modulate the social emotions guilt and shame. Serotonin depletion increased feelings of guilt and shame in individuals high in neuroticism when they were the victim of harm, whereas these emotions were attenuated when the agent of harm. These findings appear consonant with prominent work which has shown agency top-down controls the effects of stress on serotonin function, which can mitigate negative emotion, whereas the absence of agency can leave the serotonin system vulnerable to stress. The results are relevant for understanding how highly neurotic individuals may respond differently to uncontrollable stressors, have implications for their response to serotonergic medication, and may inform psychiatric classification.

Keywords: Serotonin, neuroticism, emotion, Tryptophan depletion, Depression

Disclosure: Nothing to disclose.

P235. ACCELERATED THETA BURST STIMULATION FOR TREATMENT-RESISTANT DEPRESSION IN AUTISM SPECTRUM DISORDER: RANDOMIZED SHAM-CONTROLLED TRIAL WITH EEG BIOMARKERS

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Background: Youth with Autism Spectrum Disorder (ASD) experience Major Depressive Disorder (MDD) at rates 4-fold higher than neurotypical peers, with conventional antidepressant treatments

showing limited efficacy. This treatment-resistant depression substantially increases functional impairment and suicide risk. Accelerated theta burst stimulation (aTBS), an innovative neuromodulation technique, offers promise for this underserved population. This preliminary double-blind, sham-controlled trial investigates aTBS efficacy for treatment-resistant depression in ASD while exploring novel EEG biomarkers of response.

Methods: Participants aged 13–26 years with ASD and treatment-resistant MDD (target N = 24; 12 per arm, power = 0.80, effect size $d = 1.3$, $\alpha = 0.05$, 10% dropout) are randomized to receive active or sham aTBS targeting left dorsolateral prefrontal cortex. Protocol delivers 5 sessions daily of intermittent TBS over 5 consecutive days. Primary outcome: change in Hamilton Depression Rating Scale-16 from baseline to 2-weeks post-treatment. Secondary measures include Clinical Global Impression-Improvement, suicidality, and NIH Toolbox Fluid Cognition Battery. Neurophysiological data captured via 64-channel EEG. Statistical analysis utilizes R Statistics with mixed-effects models and Bonferroni correction. Longitudinal follow-up at 2, 6, and 12 weeks post-treatment. Sex as biological variable will be analyzed.

Results: This preliminary study is actively recruiting (current N = 18). Recruitment will complete by November 2025 with full analysis available by conference date. Preliminary safety data show excellent tolerability with no serious adverse events reported. Complete efficacy, cognitive, and EEG biomarker analyses will be presented, providing first controlled evidence for aTBS in ASD-associated depression.

Conclusions: This hypothesis-generating study addresses critical treatment gaps for a vulnerable population. Results will inform clinical guidelines and advance precision medicine by identifying neurophysiological predictors of aTBS response in ASD-associated depression, potentially establishing aTBS as novel therapeutic option for treatment-resistant cases.

Keywords: Autism and depression, Non-invasive Neuromodulation, EEG biomarkers and target engagement, Cognitive/behavioral flexibility

Disclosure: Nothing to disclose.

P236. SYMPTOM-SPECIFIC AMPA RECEPTOR DENSITY PATTERNS IN BIPOLAR DISORDER AND MAJOR DEPRESSIVE DISORDER: A [^{11}C] K-2 PET STUDY

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Background: Mood disorders such as bipolar disorder (BD) and major depressive disorder (MDD) are common serious diseases that impair social functioning and lower quality of life on an individual level as well as a significant economic loss on a societal level. Although the biological mechanisms of mood disorders remain unclear, accumulating evidence suggests that synaptic dysfunction can underlie mood disorders. Excitatory glutamate α -amino-3-hydroxy-5-methyl-4-isoxazole propionic acid receptor (AMPA) is a principal component of neurotransmission. We developed [^{11}C] K-2, the first positron emission tomography (PET) tracer to quantify AMPAR density in the living human brain. Using [^{11}C] K-2, Hatano et al. reported a significant strong negative correlation between AMPAR density and the total scores in the 17-

item Hamilton Depression Rating Scale (HAMD) in the frontal lobe, while they observed a significant positive correlation between these two in the cerebellum and the part of occipital lobe such as the cuneus in patients with BD. In addition, they exhibited a significantly strong negative correlation between AMPAR density and the total scores in the HAMD in the frontal and parietal lobes in patients with MDD. Considering the heterogeneity of depressive symptoms, a detailed analysis of the symptom subdomain could provide further insights into the pathophysiology of mood disorders. This study, therefore, sought to determine whether HAMD subdomain scores correlate with AMPAR density in the brains of patients with BD and MDD.

Methods: This study comprised two clinical studies that were registered under the following IDs: UMIN000025132 and jRCTs031190150, which targeted patients with a variety of psychiatric disorders, including mood disorders (i.e., bipolar disorder and depression), and patients with mood disorders only, respectively. Data from these two studies were combined and analysed with the approval of the Yokohama City University Human Investigation Committee (trial registry number jRCT1030220303). Thirty-seven patients with BD (HAMD total score: mean $\pm$ standard deviation [SD], 6.2 ± 5.3) and 35 patients with MDD (HAMD total score: mean $\pm$ SD, 10.3 ± 7.4) underwent a PET scan with [^{11}C] K-2 and the clinical assessments. We also evaluated the three HAMD subdomains: sleep, core emotion, and atypical symptoms based on clustering analysis. The sleep domain includes energy, fatigability, midnocturnal insomnia, sleep-onset insomnia, and early morning insomnia; the core emotion domain comprises somatic anxiety, psychological anxiety, guilt and delusions, loss of interest, and mood; and the atypical domain consists of reduced libido, psychomotor slowing, suicide, psychomotor agitation, and hypochondriasis. A summed PET image at 30–50 min after injection of [^{11}C] K-2 was obtained for each participant. Standardized uptake value ratio (SUVR) images of [^{11}C] K-2 were acquired by dividing the radioactivity values of each brain region by the radioactivity value of the whole brain during 30 min and 50 min after the tracer injection (SUVR30–50 minWB). The association between SUVR30–50 minWB values and HAMD subdomain scores was assessed using multiple regression, adjusting for age and sex as covariates. Statistical significance was set at $P < 0.05$ and false discovery rate (FDR) was corrected at $P < 0.05$ (cluster-level inference, FDRc). Using SPM12, we calculated the Jaccard coefficients between the regions for the three HAMD subdomains in patients with BD and those in patients with MDD to investigate whether the detected clusters overlapped with each other.

Results: As for BD, voxel-wise analysis with SPM12 exhibited a significant positive correlation ($P < 0.05$, FDRc) between SUVR30–50 minWB of [^{11}C] K-2 PET (i.e., AMPAR density) and sleep and atypical score in the part of the occipital lobe and the cerebellum, respectively. In contrast, we observed a significantly negative association ($P < 0.05$, FDRc) between the SUVR30–50 minWB of [^{11}C] K-2 PET and core emotion score in the frontal lobe. Interestingly, these regions showed minimal overlap, suggesting that symptom-specific brain regions were distinct in BD. In MDD, the voxel-wise analysis demonstrated a significant negative correlation ($P < 0.05$, FDRc) between SUVR30–50 minWB of [^{11}C] K-2 PET and sleep, core emotion, and atypical score mainly in the orbitofrontal cortex and temporal lobe, median frontal lobe and parietal lobe, and lateral frontal and parietal lobe, respectively. These brain areas partially overlapped with each other. We identified overlapping clusters for all three symptom domains in the right frontal lobe, indicating potential core regions for all depressive symptoms. The Jaccard coefficients between the areas showing significant correlations of SUVR30–50 minWB of [^{11}C] K-2 PET with HAMD subdomains scores in BD and those in MDD were 0.006 for sleep score, 0.126 for core emotion scores, and 0 for atypical scores.

Conclusions: These findings suggest that BD and MDD are biologically distinct in terms of synaptic phenotypes. [^{11}C] K-2 PET imaging holds potential as a biological marker for differential diagnosis and symptom-specific treatment development.

Keywords: AMPA receptors, Bipolar Disorder, Major Depressive Disorder, PET Imaging, MRI

Disclosure: Viatrix, Honoraria, Self, Sumitomo Pharma, Other Financial or Material Support, Self, Wiley Japan, Other Financial or Material Support, Self.

P237. A NATIONAL 14-YEAR OBSERVATIONAL REGISTRY BASED COHORT STUDY LOOKING AT GLUCAGON-LIKE RECEPTOR 1 AGONISTS AND THEIR IMPACT ON ANXIETY, DEPRESSION, AND SUICIDALITY

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Background: People with diabetes have an elevated risk of developing depression, anxiety, and suicidality. Glucagon-like peptide-1 receptor agonists (GLP-1RA) are licensed to treat diabetes and obesity but data on whether GLP-1RA alleviate or exacerbate anxiety, depression and suicidality is mixed. We examined the rates of depression, anxiety, and suicidality in people prescribed antidiabetic medications including GLP-1RA.

Methods: We conducted an observational cohort study using national Swedish electronic healthcare registers (the National Patient registry for in-patient and specialist out-patient visits and Microdata for Analysis of Social Insurance register for sick-leave and disability pension diagnoses) to identify and follow individuals with depression (International Classification of Diseases, vol 10 (ICD-10): F32-33) or anxiety disorder (ICD-10: F40-43) through personal pseudonymized identification numbers between the years 2009 and 2022. The PRE2DUP-method was used to model medication usage by utilizing data from the National Prescribed Drug register. GLP-1RA individually and as a group were compared with non-use of GLP-1RA within the same individual, and directly with other second-line antidiabetic medications, as well as comparing them with antidepressants. To reduce bias, we used within-individual analyses where every subject was his or her own control. Rates of depression, anxiety, and suicidality in people prescribed antidiabetic medications including GLP-1RA were examined. The primary outcome was 'psychiatric decompensation' due to psychiatric hospitalisation; sick leave because of psychiatric problems; suicide attempts and death by suicide. Depression and anxiety were also separately analysed. Within-individual stratified Cox models with adjusted Hazard Ratios (aHR) and 95% confidence intervals (CIs) were used. We included 95490 individuals with 32638 of them experiencing the primary outcome and 22480 using GLP-1RA during the study period.

Results: The cohort included 60% females and had a mean age of 51 years. Use of semaglutide was associated with a 42% reduction in risk for psychiatric decompensation (aHR 0.58, 95%CI 0.51–0.65), and use of liraglutide with an 18% risk reduction (aHR 0.82, 0.76–0.89), compared with non-use of GLP-1RA, whereas exenatide and dulaglutide were not associated with psychiatric risk reduction. Semaglutide was associated with a 44% reduced risk of depressive decompensation and a 38% reduced risk of anxiety-related problems. GLP-1RA as a group were linked to reduced suicidality, and semaglutide compared favourably to other second-line antidiabetics and SSRI antidepressants.

Conclusions: Semaglutide, and to a lesser extent liraglutide, may be useful and dually effective therapeutic options, given how commonly anxiety and depression co-occur with diabetes and

obesity. Randomised controlled trials evaluating these findings are warranted.

Keywords: Anxiety and Depression, suicide, Semaglutide, GLP-1 receptor agonist

Disclosure: Johson and Johnson, Honoraria, Self, Lundbeck, Honoraria, Self, Otsuka Pharma, Honoraria, Self, Recordati, Honoraria, Self.

P238. TO BE OR NOT TO BE IMPAIRED: INDIVIDUALS WITH MILD TO MODERATE DEPRESSION DO NOT DEMONSTRATE CHARACTERISTIC DEFICIENCIES IN NEUROPLASTICITY AND SLEEP

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Background: Studies have shown that individuals with major depressive disorder (MDD) have impairments in measures of both neuroplasticity, ranging from behavioral measures of learning and memory to blood-based measures, and in sleep, such as in REM sleep. However, many of these studies have been conducted in severely depressed samples who are symptomatic despite antidepressant treatment. It is presently unclear if those who endorse milder to moderate forms of the disorder exhibit the same deficiencies. The aim of the present study was to examine measures of neuroplasticity and sleep in unmedicated individuals with mild to moderate MDD in contrast to healthy individuals.

Methods: We examined 68 unmedicated individuals (23 male) with MDD and 29 (15 male) never-depressed healthy controls (HC) across two studies investigating the effects of two unique sleep manipulations on depressive symptoms. In the morning following a baseline night of sleep in the sleep laboratory, participants completed the psychomotor vigilance task (PVT), fractal 2-back, and had blood drawn for the measurement of brain-derived neurotrophic factor (BDNF). Total sleep time, percent of each stage of sleep, and latency to sleep onset and REM were also computed. Mixed effects models were used to examine group differences.

Results: Individuals with MDD were mild to moderately depressed according to the HAM-D (Study 1: 13.38 ± 3.49 ; Study 2: 16.9 ± 6.2). Results revealed no significant differences between those with MDD and HC on the PVT, fractal 2-back, or BDNF in either study. Neither study showed decreases in REM latency or increases in percent of REM in individuals with MDD.

Conclusions: Our results demonstrated that the often-reported neuroplasticity impairments and changes in REM sleep were not found in two samples of unmedicated individuals with mild to moderate MDD. These findings may suggest that neuroplasticity impairments and certain sleep changes are associated with disorder severity. Future research should thus aim to include a broader range of participants in study samples, as more severely depressed patients are required to understand the relationships between sleep, neuroplasticity, and depression.

Keywords: Neuroplasticity, Depression, REM sleep

Disclosure: Nothing to disclose.

P239. AN EMPIRICALLY DERIVED TREATMENT ALGORITHM TO GUIDE EARLY SWITCHING OF ANTIDEPRESSANTS IN MAJOR DEPRESSIVE DISORDER: AN ANALYSIS OF THE PRIME RANDOMIZED CONTROLLED TRIAL

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Background: A substantial portion of patients with major depressive disorder do not improve with first-line antidepressants. Minimizing their length of suffering from being on a treatment that will prove to be ineffective is important. Using decision tree analysis, we constructed a decision tree that can be used to guide timely adjustments in treatment.

Methods: We analyzed 1307 patients from the Precision Medicine in Mental Health Care (PRIME) randomized controlled trial who had not attained treatment response ($\geq 50\%$ symptom improvement from baseline) after four weeks of antidepressant initiation. We focused on these participants because a clinician would not change treatment on a patient who had already attained treatment response. The participants were randomized 1:1 to pharmacogenetic testing-enhanced usual care vs. usual care alone. We conducted a decision tree analysis to determine predictors of treatment response at week-12. Baseline clinical and demographic characteristics, availability of pharmacogenetic testing results, symptom improvement at week-4, antidepressant adherence, and adverse effects were considered as potential predictors.

Results: Symptom improvement at week-4 ($X^2 = 62.0$, $p < 0.001$), better physical health ($X^2 = 9.1$, $p < 0.01$), having pharmacogenetic test results ($X^2 = 7.7$, $p < 0.01$), and not having posttraumatic stress disorder ($X^2 = 8.7$, $p < 0.01$) were predictors of week-12 treatment response. The decision tree including these predictors was able to achieve a four-fold difference in the likelihood of attaining eventual treatment response between those with the highest likelihood (rightmost terminal node in the final decision tree; $39.9 \pm 10.5\%$) vs. the lowest likelihood (leftmost terminal node, $11.1 \pm 3.0\%$).

Conclusions: Measurement based care and clinical characteristics that are easily collected in day-to-day practice can be used to identify patients for whom longer treatment on their current antidepressant is likely futile. Rather than waiting for eight more weeks, these patients can be progressed to the next step in the treatment algorithm. Future studies can apply this approach in different samples to advance personalized medicine in depression care.

Keywords: Major Depressive Disorder, Decision tree, Antidepressant

Disclosure: Nothing to disclose.

P240. RACE, CYTOKINES, AND DEPRESSION: THE ROLE OF THE EXPOSOME

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Background: Previous studies have shown that racial discrimination and the perceived stress of racism are associated with elevated pro-inflammatory cytokines. Given the established link between these cytokines and major depression - central to the inflammatory hypothesis of depression - this study examined racial differences in the inflammation-depression relationship using a secondary analysis of an ethnically diverse dataset. To extend these findings, we also conducted a post-hoc neuroimaging analysis of amygdala volume to explore potential neural correlates of inflammation across racial groups.

Methods: A total of 121 subjects were recruited as part of a larger research program on major depressive disorder (MDD) at the University of Illinois at Chicago (UIC). The sample included 46 African American participants (21 healthy, 25 with MDD) and 75 Caucasian participants (34 healthy, 41 with MDD). Depressed

participants met a minimum score of ≥ 15 on the Hamilton Rating Scale for Depression. Depression severity was also assessed using the Center for Epidemiological Studies-Depression Scale. Plasma/serum levels of C-reactive protein (CRP) and pro-inflammatory cytokines (IL-1 β , IL-6, TNF- α) were measured using enzyme-linked immunosorbent assay (ELISA) with commercially available Quantikine® kits (R and D Systems, Minneapolis, MN). Group comparisons and correlations were conducted using regression models that adjusted for demographic and neighborhood-level covariates, with post-hoc analyses examining associations between inflammation and right amygdala volume.

Results: Among non-depressed participants, African American participants had significantly higher CRP than White participants ($F = 4.48$, $p = 0.04$). No such group difference was seen among those with depression. After adjusting for neighborhood factors (e.g., household poverty, food access, unemployment), CRP differences in the non-depressed group were no longer significant ($p = 0.69$), indicating these factors may account for racial disparities in CRP.

We conducted a post-hoc neuroimaging analysis of right amygdala volume. After adjusting for age and sex, depression severity was positively associated with amygdala volume, driven largely by White participants ($r = 0.314$, $p = 0.014$), but not significant among Black participants ($r = 0.282$, $p = 0.163$). A positive association between CRP and right amygdala volume emerged in depressed Black participants ($r = 0.608$, $p = 0.016$), but not Whites ($r = 0.156$, $p = 0.232$). In non-depressed Black participants, this relationship was negative ($r = -0.328$, $p = 0.389$), suggesting better-than-expected amygdala volume given inflammation. These results underscore the need to consider social determinants of health when examining racial differences in biological correlates of inflammation.

Conclusions: The well-established association between pro-inflammatory cytokines and major depression may be moderated by race, as Black participants exhibited higher baseline cytokine levels. Among non-depressed individuals, controlling for neighborhood factors attenuated racial differences in IL-6 and CRP. Elevated IL-6 and CRP in non-depressed Black participants may reflect the cumulative impact of the exposome, providing a potential explanation for the moderating role of race in the inflammation-depression relationship. Post-hoc neuroimaging analyses further revealed racial differences in the relationship between inflammation, depression severity, and right amygdala volume: depression severity was positively associated with amygdala volume in White but not Black participants, whereas CRP was positively associated with amygdala volume in depressed Black participants. These findings highlight the importance of considering social determinants of health when evaluating both peripheral inflammatory markers and neural correlates of depression across racial groups.

Keywords: Depression Inflammation Cytokine, Neighborhood Socioeconomic Deprivation, Human Neuroimaging

Disclosure: Nothing to disclose.

P241. EXPLORING BRAIN METABOLITE SIGNATURES OF SUICIDE RISK IN TREATMENT-RESISTANT DEPRESSION: A PROTON MAGNETIC RESONANCE SPECTROSCOPY STUDY

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Background: Treatment-resistant depression (TRD) is a severe form of major depressive disorder associated with elevated suicide

risk. While structural and functional neural correlates of depression and suicide have been extensively explored, much less is known about their neurometabolic underpinnings. Addressing this gap is particularly relevant, as disruptions in neurometabolites may underlie observed anatomical and functional disruptions. In the present study, we used proton magnetic resonance spectroscopy (1H-MRS) to investigate neurometabolite alterations in the dorsal anterior cingulate cortex (ACC) in a high suicide-risk cohort of participants with TRD and a lifetime history of suicidal ideation (SI). We further examined associations between neurometabolite concentrations and suicide-related symptoms and traits, including SI severity, depression severity, anxiety symptoms, hopelessness, impulsivity and aggression.

Methods: 1H-MRS data were obtained from N = 21 participants with TRD and a lifetime history of SI, and N = 43 age- and sex-matched healthy controls (HC). Measured neurometabolites included glutamate (Glu), Glx (combined Glu and glutamine), myo-inositol, N-acetylaspartate, N-acetylaspartyl-glutamate, choline, and creatine (Cr) within the dorsal ACC. All metabolite concentrations were expressed as ratios to creatine (Cr). Metabolites showing significant between-group differences were examined for associations with suicide-related clinical and behavioral risk factors within the TRD group. Analyses were controlled for age and sex, with significance thresholded at $p < 0.05$.

Results: ANCOVA revealed that TRD participants had significantly lower Glu/Cr and Glx/Cr ratios compared to the HC group, controlling for age and sex (Glu/Cr: p -Bonferroni corrected = 0.004, partial eta squared (η^2) = 0.13; Glx/Cr: p -Bonferroni corrected = 0.043, η^2 = 0.067). Group-stratified linear regression analysis showed that within the TRD group, both Glu/Cr and Glx/Cr ratios were positively associated with total impulsivity scores (Glu/Cr: β = 0.44, p = 0.028; Glx/Cr: β = 0.48, p = 0.045), adjusting for age and sex. Additionally, Glx/Cr was positively associated with past-week SI severity (β = 0.54, p = 0.013). No significant associations were observed between Glu/Cr or Glx/Cr and aggression, depression severity, hopelessness, or anxiety symptoms. Tissue correction for gray matter, white matter, and cerebrospinal fluid fractions was explored, with no emerging significant findings.

Conclusions: 1H-MRS is currently the only non-invasive neuroimaging method capable of directly detecting and quantifying brain metabolites in vivo, yet it remains among the least utilized modalities in suicide research. This study identifies glutamatergic deficits in individuals with TRD and a history of SI in the dorsal ACC, a region implicated in emotional self-regulation and appraisal of negative experiences. Findings linking Glu/Cr and Glx/Cr to impulsivity and SI highlight a potential neurochemical target for suicide risk stratification and intervention.

Keywords: Proton magnetic resonance spectroscopy, Treatment-resistant depression, Suicide risk factors, Impulsivity, Anterior Cingulate Cortex (ACC)

Disclosure: Nothing to disclose.

P242. GLYPHALLO DOSING DOES NOT IMPAIR COGNITION IN HEALTHY VOLUNTEERS

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Background: Allopregnanolone is an endogenous neurosteroid that has antidepressant and anxiolytic activity and sleep-promoting effects through its pharmacology as a GABAA receptor positive allosteric modulator. Due to poor oral bioavailability from substantial first-pass hepatic metabolism, allopregnanolone was

previously only approved as an intravenous infusion for the treatment of postpartum depression, limiting the scope of its clinical use. A synthetic analog of allopregnanolone was previously evaluated in major depressive disorder (MDD) and showed promise but may not retain the activity, potency, and the breadth of the natural biological response to endogenous allopregnanolone.

GlyphAllo™ (SPT-300 or Glyph™ Allopregnanolone) is an oral prodrug of allopregnanolone designed to avoid first-pass hepatic metabolism by shifting absorption toward the intestinal lymphatics. GlyphAllo uses the Glyph platform, a technology which reversibly conjugates a drug of interest to a dietary fat molecule using proprietary chemistries. This route of absorption reduces side effects and enables higher oral bioavailability of allopregnanolone at a lower dose.

Central nervous system (CNS) agents, including CNS depressant drugs that target GABAergic receptors, have raised questions about potential cognitive impairment. Here, we evaluate whether GlyphAllo dosing resulted in impairment on cognition in a randomized, double-blind, placebo-controlled study in healthy volunteers.

Methods: This randomized, double-blind, placebo-controlled multiple ascending dose study included healthy male and female volunteers aged 18 to 55 years who had at least 1 post-dose baseline assessment of cognitive performance on Day 7 (NCT05129865). Subjects were randomized to receive either GlyphAllo Dose 1 (N = 7), Dose 2 (N = 9), Dose 3 (supratherapeutic; N = 9), or placebo (N = 8). Cognitive performance was assessed using the Cogstate battery, a computerized cognitive testing software that included a series of tests to measure speed of processing (SoP), attention/vigilance (AV), reasoning and problem-solving (RPS), working memory (WM), and visual memory (VM) domains. Assessments were conducted at pre-dose baseline, 3 h, 6 h, 8 h, and 12 h on Day 1 and Day 7. A composite normed z-score (mean 0, SD 1) was calculated based on average performance across normed z-scores for all 5 tasks, with a higher score indicating better performance. A normed z-score is a standardized score that indicates how many standard deviations (SD) a data point is away from the mean of a healthy normal sample.

On Day 1, the pre-dose CogState test data from healthy subjects was used to examine the reference distribution of the test scores to confirm consistency with the healthy normal sample used to derive standardized z-scores. Primary treatment comparisons were conducted using generalized estimating equations (GEE) for longitudinal data analysis. The non-inferiority margin, evaluating the treatment difference was not significantly worse than placebo, was calculated from the lower-bound of one-sided 95% confidence interval (CI) for the GEE least squares mean change in z-score from baseline to 12 h on Day 7. All p-values are nominal.

Results: A post-hoc longitudinal analysis of CogState composite normed z-score showed no-significant impairment in cognition-related parameters between any GlyphAllo dose group vs placebo. Non-inferiority bounds were within -0.18, -0.50, -0.70 SD (z-scores below healthy normal) for GlyphAllo Dose 1, 2, and 3, respectively. The 2-sided 95% CIs for between-group difference Day 7 change from baseline to hour 12 vs placebo were (-0.28, 0.93) for Dose 1, (-0.63, 0.64) for Dose 2, and (-0.84, 0.46) for Dose 3.

Analysis of CogState SoP normed z-score showed no significant difference between groups and non-inferiority bounds were within -0.43 SD (z-scores below healthy normal) across the three doses.

Analysis of CogState AV domain normed z-score showed no significant difference between groups for GlyphAllo Dose 2 and Dose 3 with non-inferiority bounds within -0.73 SD (z-score below healthy normal). The 2-sided 95% CIs for between-group difference Day 7 change from baseline to hour 12 on Day 7 vs

placebo were (−0.84, 0.68) and (−0.61, 1.12) for Dose 2 and Dose 3, respectively. Dose 1 (0.50) had a higher AV z-score than placebo (−0.48) with 95% CI (+0.27, +1.69) for Day 7 change from baseline to hour 12 ($p < 0.05$).

Analysis of CogState RPS and WM normed z-scores showed no significant difference between groups, and non-inferiority bounds were within −0.74 SD for RPS and within −0.81 SD for WM.

Analysis of CogState VM normed z-score analysis showed no significant difference between groups, and non-inferiority bounds within −1 SD (z-score below healthy normal) for GlyphAllo Dose 1 and Dose 2. GlyphAllo Dose 3 (−1.64) had a lower VM z-score than placebo (−0.45) with 95% CI (−2.27, −0.08) for Day 7 change from baseline to house 12 ($p = 0.04$).

Conclusions: In this placebo-controlled, double-blind study in healthy volunteers, there was no impairment of cognition-related parameters with GlyphAllo dosing compared to placebo. These findings suggest GlyphAllo does not impair cognition in healthy volunteers within the proposed therapeutic dose range. The BUOY-1 phase 2b study is currently underway to evaluate the efficacy and safety of GlyphAllo in participants with MDD, with or without anxious distress (NCT07065240).

Keywords: GlyphAllo, Glyph Allopregnanolone, SPT-300, Cogstate Battery, Neurosteroid

Disclosure: Seaport Therapeutics, Employee, Self, Seaport Therapeutics, Stock / Equity - Privately Held Company, Self.

P243. LOSS OF VESICULAR MONOAMINE TRANSPORTER 2 IN STRIATUM OF LONG COVID AND RELATIONSHIP TO IMPORTANT SYMPTOMS

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Background: Long COVID with neuropsychiatric symptoms is common, and effective treatments are lacking. While markers of elevated microglial and astroglial activation are elevated in the brain of Long COVID, more prominently in striatum, it is largely unknown as to whether injury to subsets of neurons occurs. Dopamine releasing nerve terminals in striatal regions may be vulnerable to injury from gliosis via excessive synaptic pruning and/or greater generation of reactive oxygen species. Moreover, dopaminergic neurons in striatum may also be vulnerable to direct infection from SARS-CoV-2 because their cell bodies in the ventral tegmental area and substantia nigra express a relatively high density of angiotensin converting enzyme 2 receptors. Here we measured (+)[11C] DTBZ binding potential (BPND), a well-established marker of dopaminergic nerve terminal integrity in the ventral striatum (VS), dorsal putamen (DP), and dorsal caudate (DC).

It was hypothesized that loss of (+)[11C]DTBZ BPND occurs in the ventral striatum, dorsal putamen, and dorsal caudate of Long COVID. Furthermore, reductions of (+)[11C]DTBZ BPND in VS, DP, and DC were hypothesized to be associated with apathy, motor retardation, and memory decline respectively. The latter is based on evidence from human illness and animal models showing that loss of dopamine neurons in these regions are associated with these particular symptoms, which are also frequent in Long COVID.

Methods: Twenty-four unmedicated, non-smoking individuals with Long COVID (12 F, 12 M; mean age 32.2) and 43 healthy controls (24 F, 19 M; mean age 29.4) underwent (+)[11C]DTBZ PET scanning. Regional (+)[11C]DTBZ BPND was quantified using a simplified reference tissue model with the cerebellum as the reference region. Group differences were assessed with linear mixed-effects model. The primary comparison was between 24 Long COVID and 24 age-matched healthy controls. A secondary comparison was between 24 Long COVID cases were compared to the larger set of similarly aged 43 healthy controls, and an exploratory comparison between 24 healthy recovered from COVID-19 and 19 healthy who never had COVID-19 (scanned prior to COVID-19 epidemic) was conducted. ROI-specific group effects were assessed using post hoc pairwise contrasts of estimated marginal means.

In addition, correlations between striatal (+)[11C]DTBZ BPND and Long COVID symptom measures of apathy, motor retardation, decline in delayed memory recall and cognitive concerns were evaluated using Pearson's correlation coefficients.

Results: For the primary analysis, (+)[11C]DTBZ BPND was reduced across all regions in 24 Long COVID compared to 24 age-matched healthy controls ($F_{1,46} = 12.7$, $P = 0.0008$). Patients with Long COVID had lower (+)[11C]DTBZ BPND in the ventral striatum (17.8%, $P = 0.006$), the dorsal putamen (15.2%, $P = 0.0004$), and the dorsal caudate (14.1%, $P = 0.003$). Moreover, (+)[11C]DTBZ BPND was reduced across all regions in 24 Long COVID compared to 43 healthy controls ($F_{1,63} = 14.5$, $P = 0.0003$). Patients with Long COVID had lower (+)[11C]DTBZ BPND in the ventral striatum (15.0%, $P = 0.006$), the dorsal putamen (13.3%, $P = 0.0002$), and the dorsal caudate (13.8%, $P = 0.005$). No significant differences in (+)[11C]DTBZ BPND were found across all regions between 24 individuals recovered from COVID-19 and 19 healthy controls never had COVID-19 (1.1% difference, $F_{1,41} = 0.21$, $P = 0.65$).

In the Long COVID group, lower (+)[11C]DTBZ BPND in the ventral striatum correlated with more severe symptoms on the Marin Apathy Evaluation Scale ($r = -0.48$, $P = 0.02$) and the Cognitive Failures Questionnaire ($r = -0.52$, $P = 0.009$). Lower (+)[11C]DTBZ BPND in the dorsal putamen correlated with slower motor speed measured by T-scores on the Finger Tapping Test Dominant Hand ($r = 0.51$, $P = 0.01$). Lower (+)[11C]DTBZ BPND in the dorsal caudate correlated with impaired memory retrieval measured by T-scores on the Hopkins Verbal Learning Test-Revised Delayed Recall ($r = 0.58$, $P = 0.003$).

These symptoms that correlated with lower (+)[11C]DTBZ BPND also differed between Long COVID cases and healthy controls who recovered well after COVID-19, as assessed by Welch's t-tests. Long COVID was associated with greater symptom severity on the Marin Apathy Evaluation Scale ($P < 0.000001$), Cognitive Failures Questionnaire ($P = 0.00005$), Finger Tapping Test Dominant Hand ($P = 0.0001$) and the Hopkins Verbal Learning Test-Revised Delayed Recall ($P = 0.00006$).

Conclusions: The most likely interpretation is that loss of dopamine synapses across ventral striatum, dorsal putamen and dorsal caudate lead to several important symptoms of long COVID. This interpretation is supported by previous electrophysiology, optogenetic, viral tracing studies as well as post-mortem studies of neurodegenerative illness which collectively demonstrate that subsets of dopamine releasing neurons in nucleus accumbens participate in motivation and cognition; subsets in dorsal putamen participate in motor control and acceleration; and subsets in caudate participate in memory retention. Hence, these findings argue for a shift in clinical trials for Long COVID towards restoring dopamine releasing synapses and/or augmenting tonic and phasic dopamine release in ventral striatum, dorsal putamen and dorsal caudate.

Keywords: PET Imaging, Dopamine, VMAT2, COVID-19, Depression

Disclosure: Nothing to disclose.

P244. REMISSION WITH LUMATEPERONE 42 MG ADJUNCTIVE TO ANTIDEPRESSANT THERAPY IN PATIENTS WITH MAJOR DEPRESSIVE DISORDER: ANALYSIS OF SHORT-TERM AND LONG-TERM TRIALS

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Background: In patients with major depressive disorder (MDD), remission rates are low ($\approx 25\%$) following first-line treatment and rates further decline with subsequent treatments. Patients who do not achieve remission have reduced quality of life and increased relapse rates, highlighting remission as a key goal of MDD treatment.

Lumateperone is an FDA-approved antipsychotic indicated in the treatment of schizophrenia and depressive episodes associated with bipolar I or bipolar II disorder. The efficacy and safety of lumateperone 42 mg adjunctive to antidepressant therapy (ADT) was demonstrated in 2 Phase 3, randomized, double-blind, placebo-controlled studies (Study 501 [NCT04985942]; Study 502 [NCT05061706]) and an open-label extension study (OLE; Study 503 [NCT05061719]) in patients with MDD with inadequate ADT response. The primary and key secondary endpoints were met in both short-term studies, with significant improvements with lumateperone 42 mg + ADT vs placebo + ADT at Day 43 in Montgomery-Asberg Depression Rating Scale (MADRS) Total score and Clinical Global Impression-Severity (CGI-S) score, respectively; efficacy was maintained throughout the OLE. Lumateperone 42 mg + ADT was generally well tolerated in both short- and long-term studies. This analysis of Studies 501, 502, and 503 evaluated MADRS Total score remission rates in patients with MDD with inadequate ADT response.

Methods: Data are presented for pooled Studies 501/502 and for Study 503. Studies 501/502 enrolled males and females (18–65 years) who met DSM-5 criteria for MDD with inadequate response to 1–2 ADTs in the current depressive episode, and had MADRS Total score ≥ 24 , CGI-S score ≥ 4 , and Quick Inventory of Depressive Symptomatology-Self Report-16 item score ≥ 14 . Patients were randomized 1:1 to 6-week, double-blind placebo + ADT or lumateperone 42 mg + ADT. Patients who safely completed double-blind treatment could enroll in Study 503 to receive 26-week, open-label, oral, once-daily lumateperone 42 mg + ADT. In pooled Studies 501/502 and Study 503, efficacy was evaluated in the overall populations and in patient subgroups (age, type of ADT, and baseline MADRS severity), using logistic regression in the placebo-controlled studies and descriptive statistics in the OLE. MADRS remission (MADRS Total score ≤ 10), complete remission (MADRS Total score ≤ 5), and sustained remission (MADRS Total score ≤ 10 at each week through end of treatment) were assessed.

Results: The modified intent-to-treat (mITT) population comprised 950 patients (lumateperone + ADT, $n = 471$; placebo + ADT, $n = 479$) in the pooled Studies 501/502; 809 patients were enrolled and treated with lumateperone + ADT in Study 503. In pooled Studies 501/502, MADRS Total score remission rates (lumateperone + ADT, 25.5%; placebo + ADT, 13.6%; $P < .0001$) were significantly greater with lumateperone + ADT vs placebo + ADT at Day 43. Remission occurred in 529 (65.4%) patients at end of open-label treatment in Study 503.

Significantly greater MADRS remission rates at Day 43 were observed in patient subgroups with lumateperone + ADT vs

placebo + ADT in the pooled Studies 501/502: younger (age ≤ 40 years: lumateperone + ADT, $n = 42$ [25.1%] vs placebo + ADT, $n = 19$ [11.6%]; $P < 0.01$), older (age > 40 years: $n = 78$ [25.7%] vs $n = 46$ [14.6%]; $P < 0.001$), selective serotonin reuptake inhibitor (SSRI) usage ($n = 91$ [28.4%] vs $n = 48$ [15.5%]; $P < .0001$), serotonin and norepinephrine reuptake inhibitor (SNRI)/other usage ($n = 29$ [19.2%] vs $n = 17$ [10.1%]; $P < .05$), baseline MADRS Total score < 32 ($n = 85$ [29.8%] vs $n = 46$ [16.0%]; $P < .001$), and baseline MADRS Total score ≥ 32 ($n = 35$ [18.8%] vs $n = 19$ [9.9%]; $P < .05$). Complete remission was achieved by 50 patients (10.6%) with lumateperone + ADT and 27 patients (5.6%) with placebo + ADT ($P < .01$) in the pooled mITT population. Sustained remission rates were significantly greater with lumateperone + ADT vs placebo + ADT beginning at Day 22: Day 22 ($n = 27$ [5.7%] vs $n = 14$ [2.9%]; $P < .05$), Day 29 ($n = 49$ [10.4%] vs $n = 28$ [5.8%]; $P < .05$), Day 36 ($n = 79$ [16.8%] vs $n = 43$ [9.0%]; $P < .001$), and Day 43 ($n = 106$ [22.5%] vs $n = 59$ [12.3%]; $P < .0001$).

In Study 503, MADRS remission rates with lumateperone + ADT at end of treatment were analyzed for subgroups based on: age (≤ 40 years: $n = 179$ [67.3%]; > 40 years: $n = 350$ [64.5%]), type of ADT (SSRI: $n = 374$ [69.9%]; SNRI/other: $n = 155$ [56.6%]), and baseline severity (baseline MADRS Total score < 32 : $n = 338$ [69.7%]; baseline MADRS Total score ≥ 32 : $n = 191$ [59.0%]). Complete remission was achieved by 357 patients (44.1%) in the OLE. Sustained remission rates were: Week 1 ($n = 107$ [13.2%]), Week 8 ($n = 231$ [28.6%]), Week 16 ($n = 301$ [37.2%]), Week 24 ($n = 330$ [40.8%]), and end of open-label treatment ($n = 346$ [42.8%]).

Conclusions: Lumateperone 42 mg + ADT demonstrated significantly greater remission rates over placebo + ADT in pooled short-term studies in patients with MDD with inadequate ADT response. Efficacy was maintained with long-term lumateperone 42 mg + ADT treatment, with ≈ 2 of every 3 patients achieving remission with 6-month treatment. These results indicate lumateperone as a promising adjunctive treatment option for patients with MDD with inadequate ADT response.

Keywords: lumateperone, major depressive disorder, remission, pooled, long-term

Disclosure: Intra-Cellular Therapies, a Johnson and Johnson Company, Employee, Self.

P245. FIRST ONSET AND DURATION OF TREATMENT-EMERGENT ADVERSE EVENTS IN PATIENTS WITH MAJOR DEPRESSIVE DISORDER TREATED WITH ADJUNCTIVE LUMATEPERONE 42 MG: A POOLED ANALYSIS OF 2 RANDOMIZED PLACEBO-CONTROLLED TRIALS

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Background: Major depressive disorder (MDD) is a common mental illness associated with functional impairment and high rates of comorbidities. Current standard-of-care treatments have adverse effects such as weight gain, sexual dysfunction, and metabolic disturbances which limit tolerability.

Lumateperone is an FDA-approved antipsychotic in the treatment of schizophrenia and depressive episodes associated with bipolar I or II disorder. Two Phase 3, randomized, double-blind, placebo-controlled studies (Study 501 [NCT04985942]; Study 502 [NCT05061706]) established the efficacy and safety of lumateperone 42 mg adjunctive to antidepressant therapy (ADT) in patients with MDD with inadequate ADT response. A pooled

analysis of these studies assessed the first onset and duration of treatment-emergent adverse events (TEAEs).

Methods: Data were pooled from Study 501 and Study 502, which enrolled males and females (18–65 years) with DSM-5–diagnosed MDD with inadequate response to 1–2 ADTs in the current depressive episode (<50% improvement on the Anti-depressant Treatment Response Questionnaire). Eligible patients had Montgomery-Asberg Depression Rating Scale Total score ≥ 24 , Clinical Global Impression-Severity Scale score ≥ 4 , and Quick Inventory of Depressive Symptomatology-Self Report-16 item score ≥ 14 . In both studies, patients were randomized 1:1 to 6-week lumateperone 42 mg + ADT or placebo + ADT. First onset and duration of TEAEs were evaluated descriptively.

Results: Of 964 patients treated (lumateperone + ADT, $n = 483$; placebo + ADT, $n = 481$), 91.4% completed treatment. TEAEs occurred in 68.1% and 45.1% of patients in the lumateperone 42 mg + ADT and placebo + ADT groups, respectively. The most common TEAEs ($\geq 5\%$ in lumateperone + ADT and twice placebo + ADT) were dizziness (lumateperone + ADT, 16.4%; placebo + ADT, 5.0%), dry mouth (12.6%; 3.3%), somnolence (10.1%; 2.1%), nausea (8.5%; 4.0%), and fatigue (7.2%; 1.2%).

Onset of TEAEs occurred at ≤ 1 week in 45.3% of patients, > 3 to ≤ 4 weeks in 4.1% of patients, and > 5 weeks in 2.1% of patients. At ≤ 1 week, > 3 to ≤ 4 weeks, and > 5 weeks, TEAE onset occurred in 11.8%, 0.7%, and 0.7% of patients for dizziness; 8.3%, 0.7%, and 0.5% for dry mouth; 8.5%, 0.2%, and 0.5% for somnolence; 6.0%, 0.2%, and 0.2% for nausea; and 5.2%, 0.2%, and 0.2% for fatigue, respectively.

The mean durations of the most common TEAEs for the lumateperone + ADT vs placebo + ADT groups were: dizziness, 9.5 vs 7.7 (median: 5.0 vs 5.0) days; dry mouth, 22.8 vs 17.3 (median: 25.0 vs 17.0) days; somnolence, 14.6 vs 14.9 (median: 9.5 vs 12.0) days; nausea, 7.2 vs 11.0 (median: 5.0 vs 6.0) days; and fatigue, 20.7 vs 11.8 (median: 19.0 vs 6.5) days.

Conclusions: In this pooled analysis of 2 studies, majority of the most common TEAEs occurred early in the treatment period and resolved approximately within 1–3 weeks of onset. These data support that lumateperone is generally safe and well tolerated as an adjunctive treatment for patients with MDD with inadequate ADT response.

Keywords: lumateperone, antipsychotic, major depressive disorder, pooled, adverse event

Disclosure: Intra-Cellular Therapies, a Johnson and Johnson Company, Employee, Self.

P246. SYMPTOM REDUCTIONS FOLLOWING 28 DAYS OF CANNABIS ABSTINENCE IN PATIENTS WITH MAJOR DEPRESSION AND CANNABIS USE DISORDER

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Background: Cannabis use disorder (CUD) presents a substantial challenge in individuals with major depressive disorder (MDD), affecting 15% of patients, a rate significantly higher than the general population (~3%). In MDD, anxiety and impairments in emotion regulation are predictive of reduced psychosocial functioning, poorer treatment response, and increased risk of suicidal behaviors. Many individuals with MDD also report using cannabis to manage sleep problems, yet few studies to date have longitudinally assessed the influence of cannabis on clinical outcomes and sleep quality in MDD, limiting causal inferences. Consequently, this study evaluates whether 28 days of cannabis abstinence improves depressive symptoms, anxiety, emotion

regulation, and sleep quality, in patients with comorbid MDD and CUD compared to continued use.

Methods: We are conducting a 4-year CIHR-funded controlled study examining the impact of 28 days of cannabis abstinence on clinical outcomes in male and female adults with MDD and CUD (target $N = 100$). Participants were randomized to contingency reinforcement (CR; abstinence reinforced with up to \$300) or non-contingency reinforcement (NCR). Weekly assessments included the Beck Anxiety Inventory (BAI), Beck Depression Inventory-II (BDI-II), Hamilton Depression Rating Scale (HAMD), Emotion Regulation Questionnaire (ERQ), and Pittsburgh Sleep Quality Index (PSQI). Linear mixed-effects models controlling for baseline substance use and demographic variables were used to assess symptom trajectories over time.

Results: To date, $N = 42$ participants have completed all study visits. Our LMMs revealed significant Time $\times$ Abstinence interactions, indicating that participants who achieved 28 days of abstinence demonstrated greater symptom improvements over the study period in anxiety (BAI, $p = 0.008$), self-reported depression (BDI-II, $p = .001$), clinician-rated depression (HAMD, $p = 0.041$), and emotion regulation (ERQ–cognitive reappraisal, $p = 0.03$) compared to those who continued cannabis use. In contrast, changes in sleep quality were not observed over the 28-day period among either groups (PSQI, $p = ns$).

Conclusions: Our preliminary findings suggest that 28 days of cannabis abstinence produces clinically meaningful improvements in anxiety, depression, and emotion regulation in adults with comorbid MDD and CUD. Conversely, abstinence was not associated with reductions in sleep quality, suggesting that short-term concerns about sleep disruption may be less pronounced than expected. Together, these results underscore the utility of targeting cannabis reduction as a complementary strategy to improve treatment outcomes in this high-risk group.

Keywords: cannabis use disorder, Major depression, emotion regulation, Anxiety, Sleep disturbances

Disclosure: Nothing to disclose.

P247. EFFICACY AND SAFETY OF BH-200, A SELECTIVE VASOPRESSIN V1B RECEPTOR ANTAGONIST, IN THE TREATMENT OF GENETICALLY DEFINED SUBGROUPS OF PATIENTS WITH MAJOR DEPRESSIVE DISORDER (MDD)

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Background: Depression is likely a biologically heterogeneous condition. A disturbance in the hypothalamus-pituitary-adrenals axis (HPA-axis) function is present in a substantial subset of depressed individuals. Several modulators of HPA-axis function (HPAMs), acting on CRHR1 or vasopressin V1b receptors (V1bR), have been investigated, but none has so far demonstrated the efficacy needed for regulatory approval. However, BH-200, a selective V1bR antagonist, showed efficacy as an antidepressant in a previous Phase 2 trial in MDD ($N = 319$). Our hypothesis is that, in a subset of depressed individuals showing a more pronounced disturbance in HPA-axis function, we may observe a more robust treatment effect. We aim to identify these patients by a genetic patient classification tool.

Methods: A genetic patient classification tool was based on the biology of vasopressin signaling and outcomes of the dexamethasone–corticotropin-releasing hormone test in depressed individuals. This tool, based on 14 single-nucleotide

polymorphisms (SNPs), classifies patients into three subgroups, A, B, and C, reflecting different levels of suppression of adrenocorticotrophic hormone (ACTH) release from the pituitary.

In the OLIVE trial, conducted in eight countries in Europe, 338 patients with MDD were randomized (2:1) to treatment with BH-200 (250 mg BID), or placebo for eight weeks. The primary efficacy endpoint was change in the 17-item Hamilton Depression Rating Scale (HAMD-17) from baseline to Week 8 in patients randomized to BH-200 and classified as belonging to Subgroup C.

Results: In the modified intention-to-treat population (mITT, N = 331), the estimated mean improvement from baseline to Week 8 was larger in the BH-200-treated arm than in the placebo arm ($\Delta = -2.98$, $p = 0.0003$). Clinically relevant improvements were observed in all three BH-200 treated subgroups A, B and C, but the magnitudes of the mean effects between the BH-200-treated arm and the placebo arm were different; Group A, $n = 89$, $\Delta = -4.47$, $p = 0.005$, Cohen's d effect size (Cohen's d), 0.55; Group B, $n = 119$, $\Delta = -2.85$, $p = 0.037$, Cohen's d, 0.49; and, Group C, $n = 123$, $\Delta = -1.94$, $p = 0.153$, Cohen's d, 0.28. The earliest separation between the BH-200-treated arm and the placebo arm was seen in Group A, where all assessments while on study drug, starting from Week 1 had a $p < 0.05$. The HAMD-17 results were supported by results on the Montgomery-Åsberg Depression Rating Scale (MADRS) at Week 8, showing an improvement in the BH-200-treated arm compared with the placebo arm in the full population of $\Delta = -5.95$, $p = 0.002$. The improvement from baseline on the Clinical Global Impression - Severity (CGI-S) scale at Week 8, comparing the BH-200-treated arm with the placebo arm was -0.46 , $p = 0.0006$ in the full population, and -0.52 , $p = 0.040$ in Group A.

The improvement from baseline, comparing the BH-200 arm with the placebo arm was maintained during the 4-week follow-up period. At Week 12, the improvement from baseline in the full population was -10.4 in the BH-200 arm, and -7.5 in the placebo arm on the HAMD-17 scale, $\Delta = -2.9$, $p = 0.0003$.

In the full population, 50.0% of the patients in the BH-200 arm and 25.7% of the patients in the placebo arm fulfilled response criteria ($\geq 50\%$ decrease in baseline HAMD-17 score) at Week 8, Odds Ratio (OR) 3.0. In subgroup A, 58.9% of the patients in the BH-200 arm and 25.9% of the patients in the placebo arm achieved response by Week 8, respectively, OR 4.2.

In the full population, 25.8% of the patients in the BH-200 arm and 16.8% of the patients in the placebo arm reached remission (HAMD-17 score ≤ 7) at Week 8, OR 1.6. In subgroup A, 33.9% of the patients in the BH-200 arm and 11.1% of the patients in the placebo arm achieved remission by Week 8, respectively, OR 3.8.

BH-200 was generally safe and well-tolerated. The most frequent adverse event was headache, reported in 8.9% of the patients in the BH-200 arm. Three serious adverse events in two patients were reported, all in the placebo arm. An elevation in liver function tests was observed in some patients. In the BH-200 arm, 5.8% of the patients had an elevation of aspartate aminotransferase (AST) or alanine aminotransferase (ALT) concentrations to >3 times upper level of normal. The changes were asymptomatic and all reversed while on treatment or after stopping the treatment.

Conclusions: We show for the first time that a drug intervention, where V1b-receptors are specifically blocked with BH-200 in an MDD subpopulation characterized by a genetic tool, results in a clinical outcome that is substantially better than in the full population. Our data confirms previous observations that HPA-axis modulators (HPAMs), such as V1b-antagonists, constitute a viable, novel class of antidepressants, and can be paired with an effective companion test. Conversely to expectations, we observed the most robust effect of the drug in subgroup A, characterized by the lowest vasopressin activity at the anterior pituitary. These results may suggest a dysbalance between the anterior pituitary and depression-relevant brain areas, enabling

BH-200 to antagonize enhanced depression-causing vasopressin-V1b signaling.

The reported ability to use a genetic tool to identify a subgroup of patients with MDD with a pronounced clinical outcome under a specific intervention, marks an important milestone in precision psychiatry. Based on the clinical data and biomarkers from this trial, the genetic patient classification tool will be further refined and used for the continued development program for BH-200.

Keywords: Precision Medicine for Depression, Vasopressin 1b antagonist, Genetic Testing, Patient Selection, Major Depressive Disorder (MDD)

Disclosure: HMNC Brain Health, Employee, Self, AstraZeneca, Stock/Equity - Publicly Traded Company, Self, H. Lundbeck A/S, Stock/Equity - Publicly Traded Company, Self, HMNC Brain Health, Stock/Equity - Privately Held Company, Self.

P248. METABOLIC PROFILES OF PARTICIPANTS WITH MAJOR DEPRESSIVE DISORDER WITH INSOMNIA SYMPTOMS IN A PHASE 3 TRIAL OF SELTOREXANT VERSUS QUETIAPINE EXTENDED RELEASE AS ADJUNCTIVE THERAPY

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Background: Currently approved medications indicated for adjunctive treatment in patients with major depressive disorder (MDD), such as quetiapine extended release (XR), belong to the class of atypical antipsychotics and have tolerability issues associated with metabolic changes or weight gain that may lead to non-adherence. Therapies in the orexin receptor antagonist class, such as seltorexant, offer a novel mechanism of action that may be a valuable alternative to atypical antipsychotic medications in the adjunctive treatment of MDD due to tolerability advantages. We examined the metabolic profiles of participants with MDD with insomnia symptoms (IS) at baseline and during a 26-week, double-blind, phase 3, randomized controlled trial (NCT04513912) of seltorexant vs quetiapine XR as adjunctive therapy.

Methods: NCT04513912 included participants aged 18–74 years with a DSM-5 diagnosis of MDD without psychotic features, an Insomnia Severity Index (patient and clinician versions) total score ≥ 15 and a positive response for IS on the Structured Clinical Interview for DSM-5 Axis I Disorders-Clinical Trials Version, and an inadequate response to 1–2 antidepressants at adequate dose and duration in the current depressive episode. Participants with Type 1 or 2 controlled diabetes mellitus at screening were eligible if otherwise medically stable, and if on a stable regimen of glucose-lowering medications for ≥ 2 months prior to screening. Participants were randomized 1:1 to receive oral seltorexant 20 mg or quetiapine XR (flexible, labeled dosage) for 26 weeks while continuing their background SSRI/SNRI. Changes in total body weight (key secondary endpoint), fasting insulin, fasting glucose, and HbA1C values were measured, along with Homeostatic Model Assessment for Insulin Resistance (HOMA-IR) score, calculated as fasting insulin $\times$ fasting glucose/22.5 (where fasting glucose >3.5 mmol/L). Low (<1.9) and high (>2.9) HOMA-IR values imply insulin sensitivity and significant insulin resistance, respectively. Body mass index (BMI, kg/m^2) categories were as follows: underweight < 18.5 , normal weight 18.5–25, overweight 25–30, and obese > 30 .

Results: Of 757 participants randomized, 756 received ≥ 1 dose of study intervention. In the seltorexant ($n = 366$) and quetiapine

XR (n = 390) groups at baseline, mean (SD) HbA1C values were 5.5 (0.6) and 5.4 (0.5); 104 (28.4%) and 107 (27.4%) participants had significant insulin resistance (HOMA-IR > 2.9); 39 (10.7%) and 27 (6.9%) participants had controlled diabetes; and 29 (7.9%) and 16 (4.1%) participants were on a stable regimen of metabolic and lipid-management pharmacotherapies. There were 125 (34.2%) and 132 (33.8%) participants who were underweight/normal in the seltorexant and quetiapine XR groups at baseline, respectively, 110 (30.1%) and 129 (33.1%) who were overweight, and 131 (35.8%) and 129 (33.1%) who were obese. The mean (SD) change from baseline to Week 26 in total body weight (kg) was 0.5 (2.9; n = 268) for seltorexant and 2.1 (3.9; n = 263) for quetiapine XR participants who received ≥ 1 dose of study intervention and had a baseline Montgomery-Åsberg Depression Rating Scale score ≥ 24 ; the least squares mean difference (95% CI) between treatment groups was -1.7 [-2.2, -1.1]; nominal $p < 0.001$). On average, all BMI subgroups gained weight at Week 26; however, mean (SD) increases in total body weight (kg) were numerically lower for seltorexant than for quetiapine XR: underweight/normal = 0.8 (2.7; n = 103) seltorexant, 2.5 (3.8; n = 90) quetiapine XR; overweight = 0.8 (2.4; n = 79) seltorexant, 2.0 (3.9; n = 91) quetiapine XR; and obese = 0.0 (3.6; n = 97) seltorexant, 1.9 (4.1; n = 93) quetiapine XR. In separate subgroup analyses of participants with diabetes, obesity, or significant insulin resistance (HOMA-IR > 2.9) at baseline, mean (SD) changes in HOMA-IR from baseline to Week 26 showed numerical decreases in seltorexant-treated participants (-3.9 [12.1; n = 22], -1.8 [8.5; n = 84], and -3.6 [9.5; n = 70]), respectively, whereas numerical increases were observed in quetiapine XR-treated participants (4.5 [12.3; n = 13], 2.2 [6.5; n = 76], and 0.2 [8.6; n = 64]), respectively. Similarly, mean (SD) changes in fasting insulin showed numerical decreases in seltorexant-treated participants (-83.4 [205.4; n = 22], -42.1 [193.2; n = 86], and -86.7 [209.5; n = 71]), whereas numerical increases were observed in quetiapine XR-treated participants (92.4 [239.5; n = 13], 61.8 [196.3; n = 77], and 22.3 [239.4; n = 65]). No clinically relevant differences in mean glucose or HbA1C levels at Week 26 were observed between treatment groups, which may be attributed to the fact that the majority of participants had baseline HbA1C values within the target range, and glycemic control was already optimized in those with diabetes.

Conclusions: Overall, body weight gain was limited after 26 weeks of treatment with seltorexant as compared with quetiapine XR. In subgroups of metabolically compromised participants, HOMA-IR and insulin levels displayed a trend towards improvement from baseline after seltorexant treatment, while a trend towards worsening was observed with quetiapine XR treatment. The majority of participants in this trial were overweight or obese and showed significant insulin resistance at baseline, highlighting an unmet need for safer treatment options in MDD.

Keywords: insomnia, Major Depressive Disorder (MDD), Metabolic side effects, Quetiapine, Tolerability

Disclosure: Johnson and Johnson, Employee, Self, Johnson and Johnson, Stock / Equity - Privately Held Company, Self.

P249. EFFICACY OF LUMATEPERONE 42 MG FOR THE TREATMENT OF MAJOR DEPRESSIVE DISORDER: ANALYSIS OF DEMOGRAPHIC AND CLINICAL SUBGROUPS IN A PHASE 3 RANDOMIZED PLACEBO-CONTROLLED TRIAL

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Background: Major depressive disorder (MDD) is a complex mental illness with varying clinical presentations and treatment responses. Certain features of patients with MDD can impact treatment outcomes. Thus, assessing treatment efficacy by patient characteristics is critical to establish consistency of treatment effect.

Lumateperone is an FDA-approved antipsychotic indicated in the treatment of schizophrenia and depressive episodes associated with bipolar I or bipolar II disorder. The efficacy and safety of lumateperone 42 mg adjunctive to antidepressant therapy (ADT) was demonstrated in 2 Phase 3, randomized, double-blind, placebo-controlled studies (Study 501 [NCT04985942]; Study 502 [NCT05061706]) in patients with MDD with inadequate ADT response. In both studies, lumateperone 42 mg + ADT met the primary and key secondary endpoints, with significant improvement vs placebo + ADT at Day 43 in Montgomery-Åsberg Depression Rating Scale (MADRS) Total score and Clinical Global Impression-Severity (CGI-S) score, respectively, and was generally safe and well tolerated. This analysis of Study 501 evaluated the efficacy of lumateperone 42 mg adjunctive to ADT in demographic and clinical subgroups of patients with MDD with inadequate ADT response.

Methods: Study 501 enrolled males and females (18–65 years) who met DSM-5 criteria for MDD with inadequate response to 1–2 courses of ADT in the current depressive episode (<50% improvement on the Antidepressant Treatment Response Questionnaire). Eligible patients had MADRS Total score ≥ 24 , CGI-S score ≥ 4 , and Quick Inventory of Depressive Symptomatology-Self Report-16 item (QIDS-SR-16) score ≥ 14 . Patients were randomized 1:1 to 6-week, oral lumateperone 42 mg or placebo adjunctive to ADT. Efficacy was evaluated in the overall population and in patient subgroups by demographic and baseline disease characteristics, using a mixed-effects model for repeated measures or analysis of covariance. Demographic subgroups included age (≤ 40 or > 40 years), sex (male or female), race (White or non-White), ethnicity (Hispanic/Latino or not Hispanic/Latino), and region (US or non-US). Baseline disease characteristics included disease severity, type of ADT, number of ADT failures in the current episode, number of lifetime ADT failures, and presence/absence of DSM-5-defined anxious distress.

Results: The modified intent-to-treat (mITT) population comprised 481 patients (lumateperone + ADT, n = 239; placebo + ADT, n = 242). Lumateperone + ADT significantly improved MADRS Total score (least squares mean difference vs placebo [LSMD], -4.9; effect size [ES], -0.61; $P < 0.0001$) and CGI-S score (LSMD, -0.7; ES, -0.67; $P < 0.0001$) from baseline to Day 43 vs placebo + ADT in the mITT population. Lumateperone + ADT significantly improved self-reported depressive symptoms at Day 43 vs placebo + ADT, as measured by QIDS-SR-16 Total score (LSMD, -2.4; $P < 0.0001$) in the ITT population.

Improvements in MADRS Total score were significant from baseline to Day 43 in demographic subgroups with lumateperone + ADT vs placebo + ADT: younger (age ≤ 40 years: LSMD, -4.6; ES, -0.57; $P < 0.001$), older (age > 40 years: LSMD, -5.1; ES, -0.64; $P < 0.0001$), male (LSMD, -4.3; ES, -0.54; $P < 0.001$), female (LSMD, -5.2; ES, -0.65; $P < 0.0001$), White (LSMD, -5.5; ES, -0.69; $P < 0.0001$), non-White (LSMD, -3.1; ES, -0.39; $P < 0.05$), Hispanic/Latino (LSMD, -9.8; ES, -1.23; $P < 0.01$), not Hispanic/Latino (LSMD, -4.5; ES, -0.57; $P < 0.0001$), US (LSMD, -5.7; ES, -0.71; $P < 0.0001$), and non-US (LSMD, -4.3; ES, -0.54; $P < 0.0001$).

Lumateperone + ADT significantly improved MADRS Total score from baseline to Day 43 irrespective of baseline disease severity (baseline MADRS Total score < 32 : LSMD, -4.5; ES, -0.56; $P < 0.0001$; baseline MADRS Total score ≥ 32 : LSMD, -5.7; ES, -0.71; $P < 0.0001$), type of ADT (SSRI: LSMD, -5.3; ES, -0.66; $P < 0.0001$; SNRI and other: LSMD, -4.0; ES, -0.51; $P < 0.01$), number of ADT failures in the current episode (1 failure: LSMD, -4.5; ES, -0.56; $P < 0.0001$; 2 failures: LSMD, -7.5; ES, -0.94; $P < 0.001$), and number of lifetime ADT failures (1 failure: LSMD, -4.8; ES, -0.61; $P < 0.0001$; 2 failures: LSMD, -4.8; ES, -0.60; $P < 0.01$). Nearly half of patients in the mITT population had anxious distress at baseline (lumateperone + ADT, n = 109 [45.6%];

placebo + ADT, $n = 98$ [40.5%]). Lumateperone + ADT significantly improved MADRS Total score from baseline to Day 43 in patients with MDD with anxious distress (LSMD, -6.8 ; ES, -0.85 ; $P < 0.0001$) and without anxious distress (LSMD, -3.5 ; ES, -0.44 ; $P < 0.001$).

Significant improvements ($P < 0.05$) in CGI-S score occurred with lumateperone + ADT vs placebo + ADT from baseline to Day 43 in all patient subgroups based on age, sex, race, ethnicity, region, baseline disease severity, type of ADT, number of ADT failures in the current episode, number of lifetime ADT failures, and baseline anxious distress. Lumateperone + ADT significantly improved ($P < 0.05$) QIDS-SR-16 Total score at Day 43 vs placebo + ADT in all patient subgroups, except for race (White, $P < 0.0001$; non-White, $P = 0.5011$).

Conclusions: Lumateperone 42 mg adjunctive to ADT demonstrated robust efficacy over placebo adjunctive to ADT in subgroups of patients with MDD based on demographic and clinical features. These results indicate lumateperone as a promising adjunctive treatment option for patients with MDD with inadequate ADT response.

Keywords: lumateperone, antipsychotic, adjunctive, major depressive disorder

Disclosure: Intra-Cellular Therapies, a Johnson and Johnson Company, Employee, Self.

P250. EFFICACY AND SAFETY OF AN NR2B NAM, BI 1569912, ADJUNCT TO BACKGROUND ANTIDEPRESSANT THERAPY IN PEOPLE LIVING WITH MAJOR DEPRESSIVE DISORDER: RESULTS FROM A PHASE II CLINICAL TRIAL

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Background: Many patients living with major depressive disorder (MDD), a common, severe, and recurrent mental illness, do not respond sufficiently to antidepressant treatment. An emerging treatment class, the NMDA receptor subunit 2B-selective negative allosteric modulators (NR2B NAMs), is hypothesized to enhance synaptic plasticity in the prefrontal cortex and thereby improve mood in MDD. BI 1569912, an oral NR2B NAM, was investigated for the treatment of MDD. Here, we describe the outcome of a Phase II, multicenter, randomized, double-blind, placebo-controlled trial in MDD patients with insufficient response to background SSRI/SNRI treatment.

Methods: 1447-0005 (NCT06280235) was an adjunctive treatment Phase II dose-finding trial that enrolled adults (18–65 years old) with an established MDD diagnosis by Diagnostic and Statistical Manual of Mental Disorders Fifth Edition (DSM-5) and insufficient treatment response in their current episode ($< 50\%$ response to 1–4 antidepressants). Participants were randomized to receive 6 weeks of treatment with either placebo or 1 of 3 doses of BI 1569912 (5 mg, 10 mg, or 20 mg) once daily in addition to their existing antidepressant therapy (randomization ratio 2:1:1:2). Assessments included the Montgomery-Åsberg Depression Rating Scale (MADRS), Symptoms of Major Depressive Disorder Scale (SMDDS), safety, and tolerability. The primary efficacy endpoint was change from baseline in MADRS total score at Week 6. Secondary endpoints included: change from baseline in MADRS total score at Day 8; treatment response ($\geq 50\%$ MADRS reduction from baseline) at Day 8 and Week 6; remission (MADRS total score ≤ 10) at Week 6; and change from baseline in SMDDS total score at Day 8 and Week 4. Dissociation was measured using the Bowdle-Visual Analogue Scale (B-VAS).

Results: Participants ($N = 243$) were randomized to placebo ($n = 79$), BI 1569912 5 mg ($n = 40$), 10 mg ($n = 41$) or 20 mg ($n = 83$). Median age (range) was 48.0 (20–65) years; 60.9% of subjects were female, and mean (SD) MADRS total score at baseline was 32.3 (5.0) points. Mean (SE) change from baseline to Week 6 in MADRS total score was -7.8

(1.6) for BI 1569912 5 mg, -11.9 (1.6) for BI 1569912 10 mg, -12.3 (1.2) for BI 1569912 20 mg and -12.0 (1.1) for Placebo. At Day 8, no relevant differences in MADRS total score were observed between treatment arms with adjusted differences (SE) for BI 1569912 vs Placebo of 1.1 (1.5) for 5 mg, 0.5 (1.5) for 10 mg and 0.7 (1.3) for 20 mg. At Day 8, no relevant difference in response rates (5 mg: 11.1%, 10 mg: 11.1%, 20 mg: 15.5%, Placebo: 11.3%) was observed. At week 6, no relevant difference in response rates (5 mg: 20.6%, 10 mg: 31.4%, 20 mg: 34.8%, Placebo: 33.8%) or remission rates (5 mg: 8.8%, 10 mg: 25.7%, 20 mg: 21.2%, Placebo: 19.7%) was observed. Mean (SE) change from baseline to Week 4 in SMDDS total score was -11.3 (1.8) for BI 1569912 5 mg, -11.8 (1.7) for BI 1569912 10 mg, -11.1 (1.3) for BI 1569912 20 mg and -10.4 (1.2) for Placebo. The most common adverse event (with incidence $> 2\%$ and higher than in placebo) in BI 1569912 total treatment group was dizziness (4.9% vs 2.5% in placebo). B-VAS scores of internal and external perception showed a difference of < 5 mm (100 mm scale) for BI 1569912 compared to placebo which is considered as not clinically relevant.

Conclusions: BI 1569912, an oral NR2B NAM investigated for the adjunctive treatment of MDD, failed to meet the primary and secondary efficacy endpoints in this Phase II trial. BI 1569912 was well tolerated with minimal adverse events and no signs of dissociation in any BI drug treated group.

Keywords: Major Depressive Disorder (MDD), adjunctive treatment, NR2B NAM

Disclosure: Boehringer, Employee, Self.

P251. A LIVING SYSTEMATIC REVIEW, META-ANALYSIS, AND OPEN DATA RESOURCE OF TRIALS OF PSILOCYBIN TREATMENT FOR SYMPTOMS OF DEPRESSION

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Background: Depression is a significant burden on health worldwide, with a growing prevalence and rising economic costs. This high burden has motivated substantial public and private investment in the development of new treatments, including treatment with psychedelics. Given the rapidly growing evidence base, systematic reviews on the topic quickly become outdated, failing to capture the most recent findings. In response, we present a living systematic review on psilocybin treatment for depressive symptoms, ensuring that the latest findings are always reflected in the data synthesis. Our review will be regularly updated; our data, code, and results will be available on our public website: SYPRES (Synthesis of Psychedelic Research Studies; sypres.io).

Methods: We included randomized controlled clinical trials published in English in peer-reviewed journals comparing psilocybin for depressive symptoms with a comparator in adult (> 18 years old) populations. We searched PubMed, Embase, PsycInfo, Web of Science, Scopus, and the reference lists of systematic reviews retrieved from the searches. Study screening and data extraction was independently performed by two reviewers. We assessed risk of bias (RoB) using Cochrane's Risk of Bias 2.0 tool. We performed inverse-variance random-effects modeling of standardized mean differences on continuous depression outcomes. Between-study heterogeneity (was calculated using the Restricted Maximum Likelihood (REML) estimator and the Q-profile method to calculate the confidence interval. We employed the Knapp-Hartung adjustment to calculate the confidence interval for the pooled effect size.

Results: Twelve studies comprising 711 participants were included in the database, with nine of those studies ($n = 529$) included in our primary model. Of the nine studies included in the primary model, two had a high risk of bias, four had some concerns, while three had a low risk of bias. Compared to control conditions, psilocybin showed a greater reduction in depression scores, with a pooled Hedges' $g = -0.91$ (95% CI, $[-1.35; -0.48]$; $k = 9$; $p = 0.0013$, $I^2 = 58.1\%$, $\tau^2 = 0.13$, $n = 501$). Sensitivity analyses revealed robust effects consistent with the primary model across a variety of design parameters and analysis choices, while also suggesting that waitlist control and crossover design studies contribute a large amount of heterogeneity to the primary model. Meta-regression revealed that psilocybin's effects were rapid and consistent over several weeks (intercept = -0.92 $[-1.26; -0.58]$, $p < 0.0001$; slope = 0.0009 $[-0.0023; 0.0041]$, $p = 0.57$).

Conclusions: This systematic review and meta-analysis suggests that psilocybin-assisted therapy results in substantial decreases in depressive symptoms across studies to date. However, many studies have small sample sizes or risk of bias. This living systematic review, meta-analysis, database, and online dashboard will continue to be updated as evidence emerges, providing a valuable resource for researchers in a rapidly evolving field.

Keywords: Depression, Psilocybin, meta-analysis

Disclosure: Nothing to disclose.

P252. IS BRAIN INSULIN ACTION IMPLICATED IN THE BIOLOGY OF DEPRESSION? A PILOT NEUROIMAGING STUDY IN ADOLESCENTS

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Background: Depression is associated with metabolic aberrations including insulin resistance, while insulin resistance is associated with greater depression severity and worse cognition. This bidirectional relationship has encouraged a reconceptualization of depression as a metabolic disorder, with brain insulin action implicated as a unifying mechanism. This has not been studied directly in humans. The aim of this study was to examine if brain insulin action is implicated in the biology of depression in adolescents.

Methods: Twelve patients with depression (both sexes), 14–18 years of age, and twelve age-, sex-, and BMI-matched healthy controls were recruited for this single-blinded crossover pilot study. To quantify insulin action in the brain, resting state function MRI (rs-fMRI) scans were performed twice: the first following intranasal placebo (INP) and the second following intranasal insulin (INI) challenge (80 IU). The difference in changes induced by INI compared to that with INP were used as an index of insulin action, since any difference observed between treatments during this controlled manipulation is likely to be due to the INI. All participants completed THINC-it, a brief cognitive assessment. The Center for Epidemiological Studies Depression Scale for Children (CES-DC) was used to assess depression severity. For cases, clinical assessments were repeated after six months. Two-way repeated measures ANOVA was conducted to assess the main effect of treatment (INI vs. INP) as within-subject factor, the participant group (cases vs. controls) as between-subject factor, and their interaction on whole-brain functional connectivity. Correlations between insulin-induced changes in rsFC with depression severity and cognition were assessed using multiple regressions.

Results: A significant group $\times$ condition interaction effect was identified for INI > INP, controls > cases contrasts. Only controls, and not cases, exhibited a significant increase in rsFC in response to INI vs. INP between the following brain regions: 1) the left Hippocampus left and Occipital Fusiform Gyrus; 2) right Parahippocampal Gyrus (PaHC-r) and left Temporal Pole (TP-l) ($F = 19.3$, $p < 0.001$); and 3) PaHC-r and right Temporal Fusiform Cortex (TFusCr) ($F = 17.5$, $p < 0.001$). At baseline, subjective cognitive performance was positively associated with changes in connectivity between the PaHC-r and TP-l ($\beta = 0.571$, $p = 0.004$); and aPaHC-r and a TFusCr ($\beta = 0.453$, $p = 0.026$). In longitudinal analyses, cases with less increase in rsFC between the TP-l and PaHC-r in response to INI vs. INP at baseline exhibited greater depression severity after 6 months ($\beta = -0.62$, $p = 0.031$).

Conclusions: This pilot study provides preliminary evidence that disrupted brain insulin action might underlie the relationship between depression and metabolic dysfunction. Replicating these findings in a larger study can implicate impaired brain insulin action early in the course of illness, which may provide us with a biological target that can be modified using insulin sensitizing interventions at the earliest stages of the illness.

Keywords: Adolescent Depression, Brain Insulin Resistance, Resting State Functional Connectivity

Disclosure: HLS therapeutics, Honoraria, Self, Boehringer Ingelheim, Honoraria, Self.

P253. THE EFFECT OF CELECOXIB ON NEUROINFLAMMATION AND DEPRESSION MEASURED BY PET IMAGING USING [18F] FEPPA

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Background: Inflammation has been hypothesized to be implicated in the pathogenesis of major depressive disorder (MDD), supported by findings of elevated levels of proinflammatory cytokines in plasma and cerebral spinal fluid (CSF) of patients with MDD. More recently, PET imaging studies have found elevated levels of neuroinflammation in brain regions implicated in MDD, quantified, in vivo, using [18F]FEPPA, a highly selective PET tracer for the translocator protein (TSPO). This study aims to investigate the efficacy of celecoxib, a cyclooxygenase-2 (COX-2) selective non-steroidal anti-inflammatory drug, in reducing neuroinflammation and depressive symptoms in patients with MDD.

Methods: Participants meeting diagnostic criteria for MDD and a current Major Depressive Episode (MDE) underwent two [18F] FEPPA PET imaging sessions, one before and one after 8 weeks of celecoxib (400 mg daily) treatment. Hamilton Depression Rating Scale (HamD) scores were obtained before and after treatment. Genotypes for the TSPO rs6971 polymorphism were collected to categorize participants as mixed affinity binders (MAB) and high affinity binders (HAB) for TSPO. Structural MRIs were obtained for region of interest (ROI) delineation. The primary ROI was the dorsolateral prefrontal cortex (dlPFC). TSPO VT (volume of distribution) was calculated from regional time activity curves using a 2-tissue compartment model previously validated for [18F] FEPPA PET and a metabolite-corrected arterial input function. TSPO VT was divided by the plasma free fraction (fP) to account for individual variations in plasma protein binding. The primary outcome measures included percent change of TSPO VT/fP and percent change of HamD scores following celecoxib treatment.

Results: In 15 MDD participants (5 MAB, 10 HAB), there was a positive association between percent change in dlPFC TSPO VT/fP and percent change in depression severity ($p = 0.006$). Change in

VT/FP explained approximately 45.6% of the variance in depression severity changes ($R^2 = 0.455$). The average HamD score for all participants decreased from 17 ± 3 at baseline to 11 ± 7 post-treatment, and 5 of the 15 participants had a final HamD score of < 7 for a preliminary remission rate of 33%.

Conclusions: Despite evidence of increased neuroinflammation in a subset of patients with MDD, there is currently no targeted approach to treat this possible underlying pathology. Our preliminary analysis suggests that celecoxib may be an effective treatment for a subset of patients with MDD and neuroinflammation.

Keywords: neuroinflammation, [18F] FEPPA, Depression, Celecoxib

Disclosure: Nothing to disclose.

P254. PRO-INFLAMMATORY MARKERS INTERACT WITH SPONTANEOUS THETA OSCILLATIONS TO PREDICT DEPRESSION AMONG ADOLESCENTS

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Background: Pro-inflammatory markers are implicated in the pathophysiology of depression, potentially altering neural oscillatory activity. Theta oscillations, especially in frontal and centroparietal regions, reflect neural processes supporting cognitive control and affect regulation, which are known to be sensitive to inflammatory signaling. This study examined whether a latent pro-inflammatory factor interacted with regional spontaneous theta power to predict depression symptoms over 12 months in adolescents, providing insight into neuroimmune mechanisms underlying symptom progression.

Methods: The sample included 190 adolescents (13–18 years old; 70% female; healthy controls = 61, remitted major depressive disorder [MDD] = 88, current MDD = 33). At baseline, participants provided passive drool samples for inflammatory marker analysis (interleukin [IL]-1 β , IL-6, IL-8, tumor necrosis factor [TNF]- α), and resting-state (eyes open) electroencephalography (EEG) data using the 10/20 electrode system were acquired. EEG data were preprocessed in EEGLAB using standard analysis pipelines, and the artifact-free signal was transformed into the frequency domain using Welch's estimation of the power spectra from 1 to 50 Hz. The Fitting Oscillations and One-Over-f (FOOOF) toolbox was used to extract periodic oscillatory power by separating oscillatory peaks from aperiodic (1/f) background activity from the resulting power spectral densities for each electrode. Spontaneous theta power was averaged across electrodes in the frontal and centroparietal topographical regions. A latent pro-inflammation factor was derived using confirmatory factor analysis, controlling for age, sex, BMI, and salivary immunoglobulin A (sIgA). Clinical follow-up assessments were completed at 6- and 12-months. A linear mixed-effects model with random intercepts was used to examine the effects of pro-inflammation, frontal theta power, centroparietal theta power, and their interactions with depression symptoms over time.

Results: The confirmatory factor analysis supported a single-factor pro-inflammation model with excellent fit, $\chi^2(14) = 18.42$, $p = 0.188$, CFI = .99, TLI = 0.98, RMSEA = 0.042, 90% CI [.000, .089], SRMR = .024. Standardized factor loadings ranged from 0.58 to 0.88 ($ps < 0.001$), and the latent factor demonstrated high internal consistency (McDonald's $\omega = .86$). Linear mixed-effects analyses revealed significant three-way interactions in which stronger frontal

theta power in the context of elevated inflammation was associated with worsening depression symptoms across 12 months ($b = 15.65$, $SE = 4.78$, $p = .001$), while stronger centroparietal theta power in the context of elevated inflammation was associated with greater symptom improvement ($b = -19.25$, $SE = 5.11$, $p < 0.001$). Of note, older age ($b = 0.13$, $SE = 0.04$, $p = 0.002$) and female sex ($b = 0.35$, $SE = 0.14$, $p = 0.015$) were significant predictors of depressive symptoms over time, whereas BMI ($b = 0.01$, $SE = 0.01$, $p = 0.423$) was not. Further, the main effects of pro-inflammation and spontaneous theta power in either the frontal or centroparietal regions alone were not significantly associated with depression symptoms over time ($ps > 0.098$).

Conclusions: These findings highlight the role of neuroimmune interactions in adolescent depression symptoms over time in which topographically specific effects suggest that spontaneous frontal theta power may index vulnerability pathways for pro-inflammation-related symptom escalation, whereas spontaneous centroparietal theta power may reflect compensatory or resilience mechanisms against elevated pro-inflammation. Integrating pro-inflammatory markers and spontaneous oscillations advances the understanding of depression pathophysiology and may inform personalized interventions targeting neuroimmune mechanisms and aid early identification of adolescents at risk for symptom escalation.

Keywords: Adolescent Depression, inflammation, Electroencephalography (EEG), Neuronal Oscillations

Disclosure: Nothing to disclose.

P255. Poster Withdrawn

P256. INTERIM RESULTS OF CONFIRMATORY EFFICACY CLINICAL TRIAL OF AMYGDALA NEUROFEEDBACK FOR DEPRESSION

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Background: Up to two-thirds of patients diagnosed with major depressive disorder (MDD) will not respond to standard pharmacological and psychological interventions, suggesting a need to develop novel therapeutics. Decreased reactivity to positive stimuli, indexed by low amygdala reactivity to positive autobiographical memory recall, may be a causal mechanism interfering with recovery from MDD. Previous work suggests that individuals who do respond to antidepressant medications show increased amygdala activity to positive stimuli that is indistinguishable from controls, while those who do not respond fail to show this increase in amygdala activity. In several small studies, we have shown that MDD participants are able to increase their amygdala response during positive memory recall via real-time fMRI neurofeedback (rtfMRI-nf) training, and that this increase is associated with large and rapid reductions in depressive symptoms. We are now evaluating the efficacy of this intervention in a large-scale confirmatory R01 clinical trial. Data collection is 50% complete and Interim results are presented here.

Methods: So far, 76 participants with MDD have completed the study. 38 were in the experimental group and received feedback regarding activity in their left amygdala, and 38 were in the yoked sham control group. Participants underwent two sessions of training where they learned to increase their amygdala response during positive autobiographical memory recall (or saw the amygdala response of another participant). Depressive symptoms were assessed biweekly via the BDI-II for 12 weeks following the final training session.

Results: The experimental group successfully increased their amygdala response with training ($p < 0.001$) while the control group did not ($p = 0.86$). With respect to clinical change, in the experimental group, BDI scores decreased an average of 41% over the follow-up period, while they decreased only 15% in the control group ($p < 0.001$).

Conclusions: While preliminary, these results suggest that we are supporting our hypotheses that patients with depression can learn to regulate hemodynamic activity from the amygdala and this ability can result in large improvements in depressive symptoms.

Keywords: real-time fMRI neurofeedback, Amygdala, Major Depressive Disorder (MDD)

Disclosure: Nothing to disclose.

P257. RESTING-STATE FUNCTIONAL CONNECTIVITY IN WOMEN WITH DEPRESSION FOLLOWING PREGNENOLONE ADMINISTRATION

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Background: Neurosteroids show promise in the treatment of mood disorders. Pregnenolone is a neurosteroid that serves as a precursor to other steroids in humans. Allopregnanolone, a GABAergic downstream metabolite of pregnenolone, is FDA-approved as an intravenous infusion (brexanolone) for the treatment of postpartum depression. Pregnenolone is available in the U.S. as a non-prescription supplement. Prior studies suggest that pregnenolone may be effective for bipolar depression. To our knowledge, no studies have examined resting-state functional connectivity (rsFC) in individuals with mood disorders following administration of exogenous neurosteroids.

Methods: Using a crossover design, women ($n = 29$) with major depressive disorder were randomized to 7 days of oral pregnenolone (500 mg/d), pregnenolone (800 mg/d), and placebo with a 14-day washout between conditions. After each treatment, serum neurosteroid levels and resting-state fMRI (rsfMRI) data were collected. A seed-based voxel-wise analysis, with bilateral amygdalae as seed, was performed to investigate rsFC alterations following treatment conditions in a per-protocol sample ($n = 11$). All included participants completed a written informed consent process approved by the UT Southwestern IRB. The study was registered at clinicaltrials.gov (NCT03645096).

Results: The mean age of participants was 46.6 (10.3) with a mean education level of 13.8 (3.7) years. A total of 24.1% were Black/African American, and 20.7% were Hispanic/Latina. About half (48.3%) were married or living with someone. The baseline 17-item Hamilton Rating Scale for Depression score was 13.9 (4.4). Compared to placebo, pregnenolone (500 mg/d) was associated with a decrease in connectivity between the bilateral amygdalae and left frontal pole ($pFDR = 0.011$). The difference in amygdala connectivity between pregnenolone (800 mg/d) and placebo was non-significant. However, compared to pregnenolone (500 mg/d), pregnenolone (800 mg/d) was associated with higher connectivity between the bilateral amygdala and left frontal pole ($pFDR = 0.015$). Pregnenolone, allopregnanolone, and pregnanolone serum levels increased several-fold with administration of either of the pregnenolone doses as compared to baseline or placebo.

Conclusions: The findings suggest that pregnenolone (500 mg/day) decreased connectivity between the amygdala and frontal pole (brain regions implicated in depression and cognitive

control). This pattern is consistent with a study by Sripada et al (2014) that observed decreased connectivity between the amygdala and dorsomedial prefrontal cortex in healthy men who received a single 400 mg dose of pregnenolone. The dose-dependent effect of pregnenolone on rsFC between these brain regions is consistent with a U-shaped response pattern. This work was supported by NIH grant R61AT009625

Keywords: Neurosteroid, Pregnenolone, Depression, Women's Mental Health, Resting State Connectivity

Disclosure: Sage/Biogen, Advisory Board, Self, Teva, Grant, Self, AbbVie, Grant, Self.

P258. EFFECTS OF INSULIN RESISTANCE, LIPID METABOLISM, INFLAMMATION AND THEIR INTERACTION ON SEVERITY OF DEPRESSION AND ANHEDONIA

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Background: Greater insulin resistance and elevated lipids have been associated with increased depression severity. Gene transcripts related to glucose and lipid metabolism have been shown to predict antidepressant response to anti-cytokine treatment. As such, metabolic disturbances may contribute to an immunometabolic phenotype of depression. In this study, we examined depressive symptoms in relation to insulin resistance, lipid metabolism, and inflammation.

Methods: 74 trauma-exposed Black women underwent assessments of insulin resistance, lipid metabolism, inflammation, and psychiatric symptoms. Depression severity and anhedonia were measured using the Beck Depression Inventory. Fatigue was measured using the Profile of Mood States. Degree of insulin resistance was determined using the Homeostatic Model Assessment of Insulin Resistance (HOMA-IR), calculated from fasting glucose and insulin levels. Lipid metabolism was determined by a composite score of summed z-transformed cholesterol (HDL negative) and triglyceride values. Inflammation was assessed by high sensitivity C-reactive protein (hs-CRP). Psychiatric symptoms were compared in participants with higher versus lower insulin resistance (based on a median split), high versus low lipid measures (based on a median split), and/or high versus low inflammation ($hs-CRP \geq 3$ mg/L versus < 3 mg/L) using t- and Mann-Whitney tests. Predictor variables were chosen based on their correlations with anhedonia. Their associations with anhedonia were examined using linear regression models with and without clinical covariates that may modify their relationships, including age, BMI, income, and education, and interaction terms.

Results: Participants with higher insulin resistance ($n = 37$) had greater depression severity (19.5 ± 9.88 vs 12.7 ± 10.2 , $p = 0.005$) and anhedonia (3 [2,4] vs 2 [0,3], $p = 0.002$) compared to those with lower insulin resistance ($n = 36$). Similarly, depression severity was higher (18.0 ± 11.5 vs 13.1 ± 8.95 , $p = 0.047$) in those with high lipid scores ($n = 36$) compared to those with low lipid scores ($n = 35$). Greater depression severity (19.1 ± 10.7 vs 11.6 ± 8.86 , $p < 0.001$) and anhedonia (3 [2,4] vs 2 [0,3], $p = 0.037$) were observed in participants with high inflammation ($n = 43$) compared to those with low inflammation ($n = 30$).

Among participants with high inflammation, there were no differences in depression severity or anhedonia between those with higher and lower insulin resistance. Within the high inflammation group, those with high lipid scores ($n = 20$) had greater depression severity (22.6 ± 11.0 vs 15.0 ± 9.26 , $p = 0.024$) and greater anhedonia (3 [2,4.5] vs 2 [1,3], $p = 0.021$) than those with low lipid scores ($n = 20$).

In the low inflammation group, participants with higher insulin resistance ($n = 15$) had greater depression severity (15.7 ± 8.92 vs 7.99 ± 6.86 , $p = 0.014$) and greater anhedonia (3 [2,4] vs 1 [0,1], $p = 0.003$) than those with lower insulin resistance ($n = 14$). There were no differences in depression severity or anhedonia between lipid score groups in those with low inflammation.

Participants with both higher insulin resistance and high inflammation ($n = 22$) had greater depression severity (22.1 ± 9.88 vs 7.99 ± 6.86 , $p < 0.001$) and greater anhedonia (3 [2,5] vs 1 [0,1], $p < 0.001$) than those with lower insulin resistance and low inflammation ($n = 14$). Similarly, participants with high lipids and high inflammation ($n = 20$) had greater depression severity (22.6 ± 11.0 vs 10.5 ± 8.11 , $p < 0.001$), anhedonia (3 [2,4,5] vs 1 [0,2], $p = 0.023$), and fatigue (4 [2,7] vs 1 [0,2], $p = 0.012$), compared to those with low lipids and low inflammation ($n = 15$).

Correlation analyses showed that insulin resistance and lipid metabolism were significantly correlated with anhedonia severity, while inflammation showed a trend towards significance. Linear regression revealed a final model in which insulin resistance, lipid metabolism, inflammation, and the interaction term between inflammation and lipids explained 30.1% of the variance in anhedonia severity ($p < 0.001$).

Conclusions: Our findings indicate that insulin resistance, lipid metabolism, and inflammation alone and in combination contribute to depression severity and anhedonia. These results support a potential role for immunometabolic mechanisms in anhedonia in a depressive subtype.

Keywords: Lipid metabolism, insulin resistance, inflammation, Depression, anhedonia

Disclosure: Nothing to disclose.

P259. TOWARD A BRIEF, SENSITIVE TOOL TO ASSESS DEPRESSIVE SYMPTOM OUTCOMES IN RESEARCH AND CLINICAL PRACTICE: LESSONS FROM THE RECOVER TRIAL

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Background: This investigation was initiated to better understand the unexpected outcomes among depressive symptom measures from the 12-month, double-blind, randomized, controlled RECOVER trial that compared active versus sham adjunctive vagus nerve stimulation (VNS) in persons with markedly treatment-resistant depression (mTRD). Total scores on the Quick Inventory of Depressive Symptomatology–Clinician (QIDS-C) and –Self-Report (QIDS-SR) were more sensitive than the Montgomery-Åsberg Depression Rating Scale (MADRS) total score in detecting overall change (baseline to 12 months across all participants) and in differentiating between the two treatment groups. Were these scales psychometrically inadequate, or were specific items insensitive to change in mTRD?

Methods: The analytic sample ($n = 463$) included all participants who had a baseline and at least one post-baseline symptomatic outcome assessment. Most patients ($436/463 = 94.2\%$) completed the 12-month randomized controlled trial (RCT). These patients had the most mTRD of any patients to ever enter an RCT, averaging over 13 prior failed antidepressant treatments (lifetime) and over 20 years in the current depressive episode. A total of 75% were unemployed, and over two-thirds had been treated with electroconvulsive therapy, repetitive transcranial magnetic stimulation, and/or ketamine/esketamine. Given the degree of treatment resistance and the known slow onset of benefit with VNS, not surprisingly, previous analyses

revealed low probability of symptomatic remission and a modest likelihood of symptomatic response, as well as the importance of partial symptom response in detecting overall change and between-group differences. Using telephone interviews, the MADRS and QIDS-C were completed by off-site, trained raters who were masked to protocol and treatment condition. Patients completed the QIDS-SR at monthly clinic visits contemporaneous with monthly MADRS and QIDS-C ratings. The Clinical Global Index of Improvement (CGI-I) was completed at the same assessment occasions by each participant's clinician (also masked to treatment assignment). All ratings were completed at baseline and monthly (months 3 to 12) after device dosing was established. To assess psychometric performance, we applied classical test theory and item-response theory (IRT) methods. We evaluated each measure for internal consistency, factor structure, test-retest reliability, item-total correlation, and the relationship of responses to each item to total score. To understand the differences in sensitivity to change, we computed the effect sizes for each item on each scale to identify the items best detecting overall change over time (across treatment groups), discriminating between the treatment groups, and detecting a partial symptom response as defined by the CGI-I.

Results: The psychometric properties of the three depressive symptom measures in this mTRD sample were acceptable and quite consistent with results from less chronically ill and treatment-resistant patients. All three measures were unifactorial with high test-retest reliability and strong internal consistency (Cronbach's alpha values above 0.8). Total MADRS scores were the least sensitive to therapeutic change both overall and to the between-treatment-group difference in symptom improvement. Item-total correlations and IRT analyses for the QIDS-C and QIDS-SR scales revealed that the sad mood, energy, interest, negative self-view, and concentration items were most strongly related to the total score. For the MADRS, these items were apparent and reported sadness, inability to feel (reflecting interest/pleasure), inner tension, lassitude (reflecting energy), and pessimistic thoughts (which include negative self-view), followed closely by concentration.

The same five items (domains) most strongly reflecting overall depression severity were identical to those that were found to be most sensitive to detecting overall change, between-group therapeutic differences, and partial response. The effect sizes of the total scores derived from the five-item versions of the MADRS, QIDS-SR, and QIDS-C were nearly identical to those of the original measures for detecting both overall outcome and between-group differences.

Conclusions: While it is widely recognized that major depressive episodes are heterogenous in phenomenology, there may be greater uniformity across patients in manifesting a common, core set of symptoms. We found that the MADRS, QIDS-C, and QIDS-SR had acceptable psychometric properties in these patients with mTRD. Five "core" depressive symptom domains—mood, interest (MADRS "inability to feel"), energy (MADRS "lassitude"), negative self-view (MADRS "pessimism"), and concentration—best reflected overall symptom severity, best identified partial responders defined by the CGI-I, and were most sensitive to detecting change over time and between-treatment-group differences. Sleep, appetite, psychomotor function, and suicidal ideation items contributed least to detecting overall benefit and between-group differences. A brief, publicly available, five-item "core" depressive symptom measure to assess treatment outcomes is feasible and applicable in both research and practice settings. Determining the optimal approach—whether clinician-rated, self-reported, or a combination of the two—deserves study.

Keywords: Psychometrics, Depression, Symptoms

Disclosure: Beckley Psytech Inc, Consultant, Self, Compass Inc, Consultant, Self, Emmes Corp, Consultant, Self, Evecia therapeutics Inc, Consultant, Self, Holmusk Technologies Inc, Consultant,

Self, ICON PLC, Consultant, Self, Johnson and Johnson, Consultant, Self, LivaNova, Consultant, Self, Mind Street Inc, Consultant, Self, Neurocrine Biosciences Inc, Consultant, Self, Otsuka-US, Consultant, Self, UT Southwestern Med Center, Royalties, Self.

P260. VAGUS NERVE STIMULATION IN MARKEDLY TREATMENT-RESISTANT MAJOR DEPRESSION: ARE THE CLINICAL BENEFITS DURABLE?

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Background: Most patients with major depressive disorder (MDD) substantially improve with one to three antidepressant treatment attempts. However, a sizable proportion (18–33%) do not, and the likelihood of obtaining initial benefit decreases with subsequent ineffective treatment steps. Critically, if initial benefit is achieved, the durability of improvement also decreases as the number of prior failed treatment trials increases. Patients who have not had sustained benefit despite multiple adequate treatment attempts have high rates of relapse, hospitalization, suicide attempt, medical morbidity, and disability. In the STAR*D trial, for example, after just two treatment failures, <5% of patients remitted and sustained their improvement at both level 3 and level 4.

The RECOVER trial tested the efficacy of adjunctive vagus nerve stimulation (VNS) compared to sham VNS in patients with markedly treatment-resistant depression. This report examined the durability of benefit (including depressive symptoms, quality of life [QoL], and daily function) achieved after 12 months of treatment with VNS combined with treatment as usual (TAU) through the second year of continued treatment. To inform clinical decisions regarding the timing of potential device explanation, we also evaluated the extent of benefit obtained after a second year of adjunctive VNS among patients with no meaningful benefit after the first year.

Methods: Participants with markedly treatment-resistant MDD received 12 months of active VNS plus TAU and continued with this combination for up to 24 months (12 months: N = 214; 18 months: N = 209; 24 months: N = 190). Patients randomized to sham VNS in the first year were excluded. Outcome domains included depressive symptom severity, quality of life (QoL), daily function, and a novel tripartite composite that combined the three domains into a single metric. The durability of benefit of each outcome domain and the tripartite metric was assessed by the proportion of participants with at least meaningful benefit (defined a priori for each domain) after 12 months who continued to sustain this degree of improvement or better at 18 and 24 months. Loss of benefit was assessed by the proportion of patients who had meaningful benefit or better at 12 months and no meaningful benefit at each follow-up. The 18- and 24-month outcomes for participants with no meaningful benefit at 12 months were examined.

Results: The vast majority of participants retained the benefit in depressive symptoms, QoL, function, and the tripartite metric observed at 12 months at 18 and 24 months. Specifically, across eight outcome measures, 78.0 to 89.2% of those with meaningful benefit at 12 months manifested this degree of improvement or better at 18 months, with a median durability value of 83.1%.

Furthermore, across outcome metrics, 79.2% to 89.6% of patients with at least meaningful benefit at 12 months had this level of improvement or better at 24 months, with a median durability value of 81.3%. The rate of loss of benefit at 18 months ranged from 10.8 to 22.0%, with a median of 16.9%. At 24 months, the rate of loss of benefit ranged from 10.4 to 20.8%, with a median of 18.8%.

Among participants with no meaningful benefit at 12 months, a substantial proportion achieved clinically meaningful benefit at 18 and 24 months. At 18 months, this characterized 23.7 to 54.7% of participants across outcome measures, with a median of 30.6%. At 24 months, the proportion with a clinically meaningful benefit ranged from 26.0 to 60.7% across outcome measures, with a median of 37.8%.

Conclusions: Approximately eight of 10 patients with a meaningful benefit in depressive symptoms, QoL, or function after one year of active VNS combined with TAU retained that benefit over the subsequent year of continued treatment. Between a quarter and half of those patients with no meaningful benefit after the first 12 months of VNS plus TAU achieved meaningful benefit during the following year of continued treatment, supporting the theory that some patients with markedly TRD require more than a year of stimulation to achieve clinical benefit. In a patient sample characterized by profound chronicity and treatment resistance, adjunctive VNS demonstrated remarkable durability of clinical improvement, and a substantial number of patients achieved meaningful clinical improvement during a second year of continued treatment.

Keywords: treatment-resistant depression, vagus nerve stimulation, durability of benefit, relapse, Major depressive disorder

Disclosure: LivaNova LLC, Advisory Board, Self.

P261. NEUROIMAGING-BASED PREDICTION OF RELAPSE AFTER ECT

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Background: Electroconvulsive therapy (ECT) remains the most effective intervention for major depressive disorder, offering rapid and robust symptom relief. Yet despite its acute efficacy, relapse is common after treatment discontinuation. Continuation ECT (C-ECT) can reduce this risk, but it brings logistical, financial, and cognitive burdens. Patients and families often push for early discontinuation once remission is achieved, even though large-scale cohort studies show that over 30% of patients are rehospitalized within a year without C-ECT—excluding additional relapses that impair function but don't result in hospitalization.

Currently, decisions about C-ECT rely heavily on clinical judgment and past treatment response, as no validated biomarkers exist to guide individualized continuation strategies. This work aims to determine whether a neuroimaging biomarker, collected near the end of the acute phase, can predict relapse during the following 6 months—directly addressing the question: Who needs continuation ECT?

Advances in longitudinal neuroimaging offer new opportunities to identify relapse risk. Depression is consistently linked to structural brain deficits, especially in the hippocampus. In our GEMRIC consortium studies, ECT has been shown to produce significant, dose-dependent increases in cortical and subcortical volumes, with the hippocampus exhibiting the most robust enlargement. Controlled studies demonstrate that greater hippocampal volume increases correlate with better clinical outcomes.

Moreover, these gains often persist for 3–6 months post-treatment, suggesting that sustained hippocampal enlargement may support ongoing stability.

Methods: We analyzed a sample of 39 patients with major depressive disorder (mean age = 51 ± 19 years; 26 female) who received a full course of electroconvulsive therapy (ECT) and were followed for a minimum of three months. Structural MRI scans were acquired at three time points: (1) baseline (pre-ECT), (2) post-acute ECT, and (3) follow-up (90–300 days after the final ECT session). Longitudinal changes in hippocampal volume were quantified using Longitudinal FreeSurfer. To characterize post-acute volume dynamics, we calculated the monthly rate of hippocampal volume change between post-acute and follow-up scans. This rate was then examined in relation to clinical outcomes, as measured by Hamilton Depression Rating Scale (HAM-D) scores at the final time point. Relapse was defined categorically as a HAM-D score ≥ 16 .

Results: Consistent with prior findings, most patients exhibited a 1–8% increase in hippocampal volume following the acute ECT course. However, hippocampal volume trajectories during follow-up varied widely: monthly rates ranged from -1.25% to $+0.25\%$. This rate of hippocampal change was strongly associated with clinical status at follow-up. Patients who relapsed showed significantly greater volume decline than those who remained in remission, both by binary classification ($t = 2.46$, $p = 0.019$, Cohen's $d = 1.49$) and by continuous HAM-D scores ($r = -0.55$, $p < 0.001$, $df = 38$). Logistic regression further confirmed this association: each one-unit decrease in volume change rate was linked to a 20-fold increase in relapse odds ($\beta = -3.02$, $p = 0.01$, $OR = 20.41$). When hippocampal volume remained stable after ECT (rate = 0), the predicted probability of relapse was only 7%. In contrast, a decline of -1.25% per month corresponded to a predicted 80% relapse risk. Notably, absolute hippocampal volume at post-acute or follow-up time points alone did not predict relapse; only the rate of volume change carried prognostic value.

Conclusions: These findings highlight the prognostic value of hippocampal volume trajectory following acute ECT. While most patients demonstrate volume increases immediately post-treatment, the subsequent rate of change - particularly during the early post-acute phase - emerges as a critical marker of relapse vulnerability. This suggests that measuring hippocampal volume within the first month of the tapering or continuation phase could provide timely, actionable information about who may benefit from ongoing continuation ECT (C-ECT). Supporting this notion, our preliminary dense-sampling data from a subset of individuals (7 subjects with ≥ 10 scans each) shows that this early decline is already detectable within the first few weeks of tapering and tracks closely with clinical outcomes. If validated in larger samples, these results could offer a mechanistically informed, individualized approach to relapse prevention after ECT.

Keywords: Depression, electroconvulsive therapy, Hippocampus, Structural MRI, relapse prevention

Disclosure: Nothing to disclose.

P262. PROPOFOL ENHANCEMENT OF SLOW WAVE SLEEP AS A PUTATIVE MECHANISM FOR TREATING DEPRESSION IN OLDER ADULTS: A PILOT STUDY

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Background: Propofol is a commonly used anesthetic whose primary mechanism of action is positive allosteric modulation of synaptic γ -aminobutyric acid A (GABA-A) receptors. Rodent models have demonstrated the ability of propofol to address homeostatic needs for non-rapid eye movement sleep. In small clinical trials, propofol has shown some promise as a novel antidepressant and enhancer of slow wave sleep (SWS) in patients with insomnia. While sleep disturbances have been recognized as a core pathophysiology of depression, it remains unknown whether promoting SWS may be important for enhancing response to oral antidepressants. We utilized propofol as a therapeutic probe in a mechanistic trial to address whether 1) propofol may augment an oral antidepressant regimen in geriatric patients with treatment-resistant depression, and 2) whether antidepressant effects are mediated through SWS enhancement.

Methods: Sixteen depressed participants, age 60 years and older who failed at least 2 oral antidepressant trials, were enrolled in an open-label trial. Participants were instructed to maintain a consistent oral antidepressant regimen throughout the study period. Two 2-hour propofol infusions were administered 2–6 days apart, with real-time monitoring to target maximal induction of slow waves on 64-channel high-density electroencephalographic (EEG). The Eleveld pharmacokinetic (PK) model was utilized for target-controlled infusions to allow stepwise titration of propofol based on the EEG, enable stable modeled brain effect-site concentrations, and post-hoc evaluation of drug exposure durations. Wireless wearable headbands were utilized to acquire at-home EEG during overnight sleep before and up to two weeks after infusions. Recordings were scored manually by AASM-registered technologists, using modified guidelines for sleep scoring of sparse frontal EEG. Depression and anhedonia were assessed using the Montgomery-Asberg Depression Rating Scale (MADRS) and Snaith-Hamilton Pleasure Scale (SHAPS), respectively. Assessments were performed within one week before the infusions and approximately 1-, 3-, and 10-weeks afterwards. Changes from baseline were calculated. Exposure durations were calculated for different ranges of modeled brain PK effect-site concentrations. Linear regressions and slope tests were utilized to evaluate associations. Nonparametric Mann-Whitney U-tests were utilized to evaluate difference in medians. The investigation was registered on ClinicalTrials.gov (NCT04680910).

Results: There were no serious adverse events and only one withdrawal. Results were based on the fifteen participants who completed both infusions. Post-infusion MADRS scores were reduced compared to baseline at 1-week (median change -4 , 95%CI $[-7,1]$, $p = 0.014$), 3-weeks (median change -5 , 95%CI $[-13,1]$, $p = 0.026$), and at 10-weeks (median change -3 , 95%CI $[-11,1]$, $p = 0.010$). Weight-adjusted average propofol dose negatively correlated with MADRS changes at 1-week ($r^2 = 0.32$, $p = 0.029$), 3-weeks ($r^2 = 0.52$, $p = 0.003$), and 10-weeks ($r^2 = 0.41$, $p = 0.010$) after the second infusion. Average change in duration of SWS on infusion nights correlated with post-infusion reductions in MADRS scores at 1-week ($r^2 = 0.27$, $p = 0.047$) and 3-weeks ($r^2 = 0.28$, $p = 0.040$). Duration of exposure at moderate modeled propofol effect-site concentrations (2.5–4.5 mcg/ml) correlated with SWS enhancement on nights following infusions ($r^2 = 0.28$, $p = 0.04$) and MADRS changes at 3-weeks ($r^2 = 0.42$, $p = 0.009$). In contrast, exposure at low modeled propofol effect-site concentrations (1–2 mcg/ml) did not correlate with SWS changes ($r^2 = 0.21$, $p = 0.08$), instead negatively correlated with MADRS improvements ($r^2 = 0.37$, $p = 0.016$). In evaluating anhedonia, changes in SHAPS relative to baseline at 1- and 3-weeks were not significantly different from 0 (median changes 0 and 1, respectively, with both $p > 0.05$). In contrast, at 10-weeks post-infusion, there was a small but significant improvement (median change: -2 points,

$p = 0.011$) not associated with either weight-adjusted dose or duration of exposure at modeled propofol effect-site concentrations (2.5–4.5 mcg/ml, both $p > 0.05$).

Conclusions: These open-label trial findings suggest that serial propofol infusions may augment oral antidepressant therapy in geriatric patients with treatment-resistant depression, in a dose-dependent manner. Enhancement of post-infusion SWS is a potential mechanism for subsequent antidepressant response. These safety, feasibility, and response data support an ongoing experimental double-blind randomized controlled trial (ClinicalTrials.gov NCT06867549).

Keywords: NREM sleep, Propofol, GABAA receptor positive allosteric modulator, Electroencephalography (EEG), Late-life Depression

Disclosure: Nothing to disclose.

P263. SEX DIFFERENCES IN PLACEBO AND ANTIDEPRESSANT RESPONSE TO INTRANASAL ESKETAMINE FOR TREATMENT-RESISTANT DEPRESSION: A POOLED PARTICIPANT ANALYSIS OF RANDOMIZED CONTROLLED TRIALS

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Background: Racemic ketamine and its enantiomer, esketamine, have emerged as fast-acting antidepressant options for individuals with treatment-resistant depression (TRD). Yet, despite growing clinical use, little is known about how sex assigned at birth shapes symptom-specific responses to these interventions, a critical gap in the move toward personalized psychiatry.

Methods: We conducted a pooled analysis of 5 randomized, double-blind, placebo-controlled trials in which adults with TRD received intranasal esketamine or placebo twice weekly for four weeks, alongside a newly initiated oral antidepressant for over 1000 patients. We evaluated the effects of sex assigned at birth on overall depression severity, measured via total Montgomery-Åsberg Depression Rating Scale (MADRS) scores, and across four symptom factors: sadness, negative thoughts, detachment, and neurovegetative symptoms. Rates of clinical response and remission were also analyzed by sex assigned at birth.

Results: Overall, esketamine treatment improved total MADRS scores in both sexes; however, significant sex-specific patterns emerged. Females showed greater improvement in total MADRS scores than males towards the end of the trials, in both the placebo and esketamine arms. Females also showed more pronounced reductions in the sadness and detachment factors at the end of the trials, as well as in the neurovegetative factor on day 15, regardless of the treatment group. On the other hand, males showed a significant reduction in sadness symptoms after esketamine on day 2 of the treatment. Females had higher odds of responding, regardless of treatment arm, during later time points.

Conclusions: These findings reveal that sex assigned at birth influences overall antidepressant response and shapes the trajectory and symptom profile of improvement. Our findings emphasize the critical importance of incorporating sex assigned at birth as a key variable, essential for optimizing TRD treatment strategies and advancing individualized mental healthcare.

Keywords: Esketamine nasal spray, Major Depression Disorder, MADRS, sex differences, remission

Disclosure: Nothing to disclose.

P264. CEREBRAL ORGANOID FROM TREATMENT-RESISTANT DEPRESSION PARTICIPANTS AND HEALTHY VOLUNTEERS DISPLAY STARK DIAGNOSTIC DIFFERENCES AND DISPARATE TREATMENT RESPONSES

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Background: Treatment-resistant depression (TRD) is a pervasive neuropsychiatric disorder that worsens long-term outcomes, promotes frequency of recurrent episodes, and amplifies suicidal ideation [1]. Despite decades of psychological and pharmacological research, breakthrough treatments for TRD have remained elusive, with a rare exception the emergence of subanesthetic doses of ketamine. (2R,6R)-hydroxynorketamine (HNK) is a major ketamine metabolite under investigation as a rapid-acting antidepressant without dissociative effects [2]. However, its exact mechanism of antidepressant action is still unclear. The inability to assess brain-specific molecular changes in clinical populations have limited our understanding of how novel effective antidepressants, such as ketamine its metabolites, are impacting molecular neurobiology in depression. Induced pluripotent stem cell (iPSC)-based modeling, particularly the generation of cerebral organoids derived from patient-donated samples, has provided a novel avenue to assess these changes within neuropsychiatric disorders [3].

Methods: Peripheral blood mononuclear cells were collected from participants in a randomized controlled clinical trial at the NIMH, conducted at the NIH Clinical Center in Bethesda, MD, USA (NCT02484456). All provided written informed consent before taking part in this study, which was approved by the NIH Institutional Review Board. TRD participants were operationally defined here as a history of inadequate response to at least one conventional antidepressant treatment, using the modified-Antidepressant Treatment History Form. The PBMCs were collected at baseline and reprogrammed using non-integration methods to iPSCs at the National Heart, Lung, and Blood Institute (NHLBI/NIH) iPSC Core Facility. All participants in this subsample were female (TRD: $n = 5$, healthy volunteers (HV): $n = 5$). Cerebral organoids were generated following STEMCellTM protocols and aged for 60 days to ensure maturation and diversity of cell types. At 60 days, organoids were exposed to either (2R,6R)-hydroxynorketamine (HNK) ($1\mu\text{M}$) or vehicle, which was washed out after one hour. Organoids were collected for bulk RNA-sequencing analyses after 24 hours. DeSeq2 determined differential gene expression, which was then analyzed using Ingenuity Pathway Analysis (IPA, Qiagen). A log fold change ≥ 1.2 was used to filter the dataset and all p-values presented are Benjamini-Hochberg (BH) corrected.

Results: Utilizing the IPA Diseases and Functions database, TRD cerebral organoids demonstrated a significant overlap (12.23%, $p = 7.64\text{e-}8$) with previously published depressive disorder gene sets, confirming a depressive-like phenotype within this in vitro model. Additionally, a significant overlap (33.33%, $p = 6.24\text{e-}5$) with known molecules involved in TRD was found. Compared to HV cerebral organoids, significant decreases were found in phagosome formation ($p = 4.61\text{e-}11$, $z = -3$), S100 signaling ($p = 4.61\text{e-}11$, -0.83), neuronal CREB signaling ($p = 3.81\text{e-}10$, $z = -0.72$), and G-alpha (i) signaling events ($p = 7.96\text{e-}6$, $z = -1.47$). (2R,6R)-HNK treatment increased IGF1 ($p = 0.0057$, $z = 3.302$) and EGF signaling ($p = 0.001$, $z = 2.351$) in TRD organoids. Paralleling previous findings in iPSC-derived cortical neurons, TRD organoids also demonstrated increases in upstream regulators of inflammatory signaling including tumor necrosis

factor (TNF) ($p = 1.45 \times 10^{-12}$, $z = 4.29$), IL-1B (1.42×10^{-10} , $z = 3.48$), and IL-13 ($p = 3.49 \times 10^{-10}$, $z = 1.81$). HV organoids were not responsive to (2R,6R)-HNK and showed no changes in signaling pathways or molecules that survived BH correction (corrected $p > 0.05$).

Conclusions: By utilizing an innovative iPSC-based cerebral organoid model, we have effectively mimicked the depressive phenotype and delineated distinct molecular signatures associated with TRD, such as decreases in neuronal CREB and G-alpha (i) signaling events [4,5]. The enhancement of IGF1 signaling in TRD organoids following (2R,6R)-HNK treatment aligns with preclinical data supporting a role for IGF1 release in the medial prefrontal cortex in ketamine's antidepressant effects [6]. The lack of response in HV organoids suggests a specificity of (2R,6R)-HNK's action to pathological states, paralleling clinical research findings in ketamine [7]. These data validate the use of cerebral organoids as a viable non-animal model for studying neuropsychiatric disorders and highlight the therapeutic promise of (2R,6R)-HNK through molecular mechanisms shared with racemic ketamine.

Keywords: Brain Based Markers for Depression, Brain organoids, (2R,6R)-hydroxynorketamine, Treatment-resistant depression

Disclosure: Nothing to disclose.

P265. MIR1255A REGULATES PATHWAYS CRITICAL FOR BRAIN DEVELOPMENT, RISK GENES FOR DEPRESSION AND NEURODEVELOPMENTAL DISORDERS

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Background: Multiple lines of evidence implicate microRNAs (miRNAs) in major depressive disorder (MDD), including the location of miRNAs genes (MIRs) within genomic regions identified by genome wide association studies (GWAS).

Methods: To investigate the role of MIRs in the genetic risk for MDD, we identified all MIRs in GWAS linkage disequilibrium (LD) regions for MDD. We then identified the associated single nucleotide polymorphism (SNPs) that were expression quantitative loci (eQTLs) for these MIRs in brain. Based on pathway analyses of the predicted targets, we selected MIR1255A for functional studies and overexpressed it in human neural precursor cells (NPCs) to identify target genes in neural cells.

Results: Our analyses identified 107 MIR genes within MDD LD regions, with four of these having associated eQTLs in the credible SNP set (variants near a genetic association signal predicted to include causal variants). Transcriptome analyses of the MIR1255A transfected NPCs identified 370 differentially expressed (DE) genes at an adjusted p (FDR) < 0.05 and an additional 249 DE genes at adjusted $p < 0.1$ (total 619 genes). Of these, 85 genes were in genomic regions identified by MDD GWAS, with some supported as risk genes from biological studies (altered expression or serum levels in individuals with MDD, animal models). Notably, 157 DE genes were implicated in neurodevelopmental disorders. Gene set enrichment analyses of DE genes revealed the top categories as neurogenesis, generation of neurons, regulation of cell differentiation and neuron development.

Conclusions: These findings support MIR1255A as regulator of processes critical for neurodevelopment and a contributor to genetic risk for depression.

Keywords: MicroRNA, gene transcription, Genetics of depression, neural progenitor cells

Disclosure: Nothing to disclose.

P266. SIMULATED TRADEOFFS BETWEEN CONTINUOUS AND EPISODIC ANTIDEPRESSANT TREATMENT IN RECURRENT DEPRESSION

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Background: Clinical guidelines recommend indefinite antidepressant maintenance treatment (ADM) for patients with three or more depressive episodes. Yet many patients have concerns about continuous ADM use, citing side effects such as sexual dysfunction or emotional blunting, or a preference to avoid long-term dependence. For such patients, episodic treatment during relapses—with active monitoring in between—may be a more acceptable alternative, depending on their personal balance of benefits and harms. This approach reduces total ADM exposure but increases time spent with depressive symptoms due to delays in treatment initiation and the slow onset of medication efficacy. To inform patient decision-making, this tradeoff must be quantified in a direct, like-to-like comparison—something not previously modeled. We therefore performed hypothesis-generating simulations the long-term consequences of continuous versus episodic treatment with active monitoring in recurrent major depressive disorder.

Methods: We introduced a new measure of effect size, Days Needed to Treat (DNT), a within-person analogue of Number Needed to Treat, representing the additional days of medication exposure (for continuous vs. episodic treatment) required to avert one day of depression. We conducted simulations in R to estimate this measure under varying clinical scenarios. Patients were simulated over 20 years, experiencing relapses at frequencies ranging from once every 2 years to once every 10 years. Episode durations ranged from 3 to 11 months, consistent with prior estimates (Spijker 2002; ten Have 2017). In the core model, ADMs reduced depressive symptom intensity by 50% at full strength (Stone 2022), whether used for prevention or treatment (symptom-masking hypothesis; Hollon 2005). Effectiveness ramped to 71% of full strength by week 4 and 100% by week 12 (Henssler 2018). Under episodic treatment with active monitoring, episodes were assumed to be recognized and treatment restarted within 4 weeks (range 2–6), continuing until episode resolution. To account for parameter uncertainty, we ran 10,000 probabilistic simulations per relapse-frequency category. Standard deviations reflected plausible ranges and were tested in sensitivity analyses. Statistical significance testing was not performed, as p -values would only reflect the number of simulation runs. Sex as a biological variable did not play a conceptual role in the simulation model but would be expected to be assessed in any future empirical studies validating these simulations.

Results: Across 10,000 simulated patients per recurrence frequency scenario, each depressive episode under episodic treatment produced a mean of 7.5 (95% CI: 4.7–10.6) additional effective weeks of untreated depression compared to continuous treatment. After adjusting for the partial efficacy of ADMs, this corresponded to 3.7 (1.9–6.1) additional weeks lived with depression per episode. For patients with moderately recurrent depression (one episode every 5 years; ~4 episodes over 20 years), episodic treatment led to 104 (23–241) additional days of depression but 6559 (5434–7178) fewer days of ADM exposure. This yielded a DNT of 91.7 (25.1–315.3): on average, 3 months of ADM exposure were required to prevent one day of depression. For patients with other recurrence frequencies (every 2, 3, or 10 years), the DNT was 24.7 (9.0–56.7), 44.0 (14.9–115.1), and 184.4 (42.8–460.6), respectively.

Conclusions: Our simulations suggest that a patient with recurrences once every 5 years would prefer continuous treatment over episodic treatment with active monitoring only if they value avoiding 1 day of depression more than avoiding 3 months of ADM exposure. Different tradeoffs emerge for patients with more or less frequent recurrences. Although parameter values were drawn from the literature, these findings are based on simulations and are valid only insofar as the model reflects reality and these results should be viewed as hypothesis-generating until reproduced with patient data. More broadly, the results highlight the possibility that episodic treatment with active monitoring may be a value-concordant option for patients who struggle with ADM side effects or who experience infrequent recurrences.

Keywords: Depression, relapse prevention, value-based decision making, Simulation

Disclosure: Nothing to disclose.

P267. DEVELOPMENT AND EVALUATION OF AN $\alpha 5$ SUBUNIT-CONTAINING GABA-A RECEPTOR SELECTIVE NAM AS A RAPID-ACTING ANTIDEPRESSANT WITHOUT THE UNDESIRABLE CNS EFFECTS OF KETAMINE AND PSYCHEDELICS

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Background: Esketamine, the S-enantiomer of ketamine, is the first FDA-approved rapid-acting treatment for treatment-resistant depression (TRD) and major depressive disorder with suicidal ideation or behavior (MDSI). Ketamine and its enantiomers are potent neuroplastogens that rapidly induce neurotrophic signaling, promote synaptogenesis, and reverse stress-induced synaptic deficits. At the circuitry level, ketamine restores the balance between excitation and inhibition in circuits regulating mood and reward. However, as non-subtype-selective NMDA receptor antagonists, ketamine and its enantiomers produce undesirable CNS effects—including dissociation, transient cognitive impairment, and abuse liability—that limit their clinical utility.

The $\alpha 5$ subunit-containing GABA-A receptor ($\alpha 5$ GABAAR), unlike other GABA-A receptor subtypes, does not mediate the anxiolytic, sedative, or antiepileptic effects of GABA or benzodiazepine-like drugs. Selective antagonists and negative allosteric modulators (NAMs) of $\alpha 5$ GABAARs, such as basmisanil, were originally developed primarily for cognitive enhancement. Both animal and human studies have shown that these agents do not produce the undesirable CNS effects associated with ketamine and psychedelics, and with subtype non-selective GABA-A receptor NAMs such as flumazenil. In chronic stress models of depression, these agents ameliorate anhedonic behaviors within 24 h of administration and are also effective in other depression models and antidepressant-sensitive behavioral tests. Furthermore, this class of agent induces neurotrophic signaling and restores synaptic plasticity similar to the effects of ketamine.

The current study aims to develop and evaluate PNV001, a metabolically improved $\alpha 5$ GABAAR NAM, in preclinical studies as a basis for a future proof-of-concept clinical trial for depression.

Methods: Human brain single-cell gene expression data were obtained from the Allen Brain Institute. Only cells that passed quality-control requirements were included for examining the expression of GABA-A receptor subunits in selected brain regions and cell types. Basmisanil was purchased, and PNV001, a deuterated analog of basmisanil at a key metabolic breakdown site, was synthesized. GABA-A receptor activities of basmisanil and PNV001 were measured using manual patch-clamp assays with cell lines expressing recombinant human GABRA1, GABRA2,

GABRA3, and GABRA5 together with GABRB2/3 and GABRG2. Cross-species liver microsome stabilities were determined using well-established methods. Anti-immobility effects and reversal of anhedonia-like behaviors in stressed animals were assessed as in previously reported studies.

Results: The $\alpha 5$ subunit is detectable in excitatory neurons of the cerebral cortex (58%), hippocampus (79%), and amygdala (69%), as well as in dopaminergic (17%) and serotonergic (22%) neurons of the midbrain. Compared with basmisanil, PNV-001 maintains the desired potency at $\alpha 5$ GABAARs ($IC_{50} = 2-3$ nM) and demonstrates high selectivity over other 5GABAAR subtypes (>100-fold). PNV-001 also shows marked improvements over basmisanil in half-life in human (5.5-fold), monkey (8.1-fold), and dog (2.4-fold) liver microsomes. Similar to tool compounds and basmisanil, PNV-001 significantly reduced immobility in the tail suspension test ($F(2,9) = 8.03$, $p = 0.01$) and forced swim test ($F(2,9) = 19.56$, $p = 0.0005$) rapidly and persistently (24 h), and restored sucrose preference ($F(2,9) = 5.87$, $p = 0.023$) and social interaction ($F(2,9) = 10.61$, $p < 0.004$) tests within 24 hours in rats rendered anhedonic by chronic stress.

Conclusions: These data indicate that PNV-001 has a promising profile as a mechanistically novel, first-in-class, rapid-acting treatment for TRD and MDSI, without the undesirable CNS effects associated with ketamine and psychedelics.

Keywords: Antidepressant, TRD, (R,S)-ketamine, Psychedelics, GABA-A receptors

Disclosure: ProNovo Therapeutics, Founder, Self.

P268. EFFECTS OF ELECTROCONVULSIVE SHOCK ON THE HIPPOCAMPAL TRI-SYNAPTIC CIRCUIT

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Background: Therapeutic use of electroconvulsive shock (ECS) therapy is particularly effective for the remission of treatment-resistant depression. Yet the question of how ECS produces these powerful effects remains unanswered. Like other more common forms of antidepressant treatment such as fluoxetine, ECS has been shown to increase neurogenesis in the hippocampal dentate gyrus of rodent models. Yet the question of how ECS-induced neurogenesis supports improvement of depressive symptoms remains unknown. Here we use a mouse model to examine the effects of ECS on immature vs mature granule neurons as well as the principal neurons of the tri-synaptic circuit. First, we test the hypothesis that the presence of immature granule neurons is necessary for the therapeutic effects of ECS. We then use slice electrophysiology to ask how activity in immature granule neurons affects activity of mature granule neurons in ECS vs Sham subjects. Using single nucleus RNA sequencing, we then transcriptomically profile CA1 and CA3 pyramidal cells as well as dentate gyrus granule neurons after ECS vs Fluoxetine treatment.

Methods: Male mice were exposed to chronic corticosterone to induce a stressed phenotype. ECS was delivered every other day for 10 sessions in the experimental group, while Sham mice received isoflurane anesthesia and ear clip placement identical to the ECS group, yet no current was delivered to Sham mice. To ablate neurogenesis, X-irradiated mice received three sessions of x-ray exposure targeting the hippocampus bilaterally, while Sham mice received anesthesia only ($N = 10$ /group). For slice electrophysiology, we used a tamoxifen inducible nestin-creERT2 line crossed with a cre-dependent channelrhodopsin (ChR)-YFP to allow us to specifically activate immature granule neurons ($N = 5$

per group). We then measured electrophysiological responses in mature granule neurons. For RNA-sequencing experiments ($N = 3$ per group), we sorted for NeuN expressing cells in order to limit our exploration to neurons, and analysis was performed using Seurat. Antidepressant-like behavior was assessed by the novelty suppressed feeding test and the forced swim test.

Results: We found that that ECS-induced neurogenesis is necessary to improve depressive-like behavior of mice exposed to chronic corticosterone. Using slice electrophysiology, we found that optogenetic stimulation of immature granule neurons produces a hyperpolarization in mature granule neurons. Consistent with this finding, we observed reduced expression of the early immediate gene cFos in ECS vs sham subjects (Student's T test; $p = 0.0007$). This result indicates that ECS amplifies the effect of a direct, inhibitory connection between immature and mature granule neurons. Using single nucleus RNA sequencing, we reveal major transcriptomic shifts in granule neurons after treatment with ECS+Cort or fluoxetine+Cort vs Cort alone. We identify a population of immature-like cells which has greater representation in both ECS+Cort and fluoxetine+Cort treated samples vs Cort alone (one-way ANOVA, $F(3, 8) = 11.00$, $p = 0.0044$). We also find global differences in ECS- vs fluoxetine-induced transcriptomic shifts in both granule neurons and CA1 and CA3 principal neurons.

Conclusions: Together, our findings contextualize the role of immature granule neurons as mediators of the therapeutic effects of ECS. We show that ECS increases the ability of immature granule cells to suppress activity of mature granule cells. We then reveal the impact of ECS beyond granule neurons to shape expression in both CA1 and CA3 pyramidal cells.

Keywords: Hippocampal neurogenesis, electroconvulsive therapy, RNAseq, Hippocampus, Antidepressants

Disclosure: Nothing to disclose.

P269. BIOLOGICAL RHYTHMS DISTURBANCE AS A PREDICTOR OF DEPRESSION: FINDINGS FROM AN ANIMAL MODEL

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Background: Recent studies have exposed a notable relationship between the inadequate functioning of biological rhythms and the emergence of mental disorders, especially major depressive disorder (MDD). Changes in biological rhythms cause sleep disorders, which are one of the most common symptoms in individuals with MDD.

Methods: This study aimed to investigate the behavioral and neurochemical effects of male and female rats subjected to the altered biological rhythms (ABR) protocol. For this, we used Wistar rats, 60 females (F) and 60 males (M), which were divided into two groups: control and RBA. The RBA group was subjected to a protocol of changes in adequate sleeping and feeding models for 40 days. Afterward, the rats underwent behavioral tests: depressive-like behavior, spontaneous motor activity, habituation memory, anhedonia, and anxiety-like behavior. Serum, prefrontal cortex, and hippocampus samples were collected to measure the levels of the cytokines interleukin-1 β (IL-1 β), interleukin-6 (IL-6), and interleukin-10 (IL-10), as well as oxidative stress parameters, including carbonyl, lipid damage, superoxide dismutase (SOD), and catalase. The prefrontal cortex, hippocampus, and hypothalamus were used for the analysis of the clock genes CRY2 and PER2 through western blotting.

Results: The results demonstrated that the ABR protocol was capable of promoting anhedonic, depressive, and anxious

behavior, and caused cognitive damage ($p < 0.05$) in male and female rats that underwent this protocol. In the hippocampus of females in the RBA group, there was an increase in the levels of IL-1 β , IL-6, and IL-10, as well as SOD activity ($p < 0.05$), compared to the control group. In the hippocampus of males, SOD activity was reduced in the RBA group compared to the control ($p < 0.05$). A decrease in CRY2 and PER2 levels was found in the hypothalamus and prefrontal cortex of male and female rats. Furthermore, an increase in PER2 was observed in the prefrontal cortex of female rats ($p < 0.05$).

Conclusions: These findings suggest that changes in biological rhythms may be a prodrome for the development of MDD and its comorbidities. The RBA protocol was able to alter clock genes, inflammatory parameters, and SOD in the hippocampus, suggesting an association with the effects of changes in biological rhythms, especially in females.

Keywords: biological rhythms, neuroinflammation, psychiatric disorders

Disclosure: Nothing to disclose.

P270. SEX DIFFERENCES IN DOPAMINE D1-D2 RECEPTOR HETEROMER FUNCTION MEDIATES DIVERGENT MECHANISMS OF DEPRESSION SUSCEPTIBILITY

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Background: The biological mechanisms driving the 2-3-fold higher incidence of depression in women compared to men remain unclear, resulting in significant unmet patient needs and highlights the need for novel antidepressants that effectively reduce MDD burden in both sexes. Activation of the brain dopamine system is traditionally linked to positive reward regulation, which becomes blunted in MDD to confer hallmark symptomatology of low mood and anhedonia. In contrast, activation of the dopamine D1-D2 receptor heteromer (D1-D2 heteromer) precipitates behavioural aversion via G α_q -mediated calcium signaling cascades, Ca $^{2+}$ /calmodulin-dependent protein kinase II α (CaMKII α) activation, brain-derived neurotrophic factor (BDNF) and prodynorphin (PDYN) production. Compared to male animals, the striatum of female rat and primate harbored higher basal density of D1-D2 heteromer. Female rats required subthreshold heteromer activation to precipitate anhedonic-, depressive-, and anxiety-like behaviours. Given that D1-D2 heteromer activation promotes aversion and curtails reward — processes exaggerated in human MDD — D1-D2 heteromer function may be enhanced in a parallel “anti-reward” dopaminergic neural circuit to confer sex-specific depressive symptoms. The present study investigated D1-D2 heteromer-mediated mechanisms of depression susceptibility unique to male and female rat in a chronic unpredictable stress (CUS) model of MDD.

Methods: Adult male and female Sprague-Dawley rats were exposed to 2–3 daily, randomized mild stressors for 24 days (cage tilt, wet bedding, bright lights, food/water deprivation, forced swim, tube restraint); control animals were handled 4 times weekly. A selective interfering peptide targeted to the D1-D2 heteromer interaction site was administered via intranasal route acutely prior to behavioural testing, including sucrose preference (days 1, 8, 15, 21), elevated plus maze, and social interaction tests. Serum corticosterone was evaluated by enzyme-linked immunosorbent assay 1 h after stress exposure on days 7 and day 24. Prefrontal cortex, striatum, and amygdala tissues were collected 1 hour after the last stressor on day 24 for evaluation of D1-D2 heteromer density by proximity ligation assay (PLA) and fluorescent in situ hybridization. Activation of D1-D2 heteromer signaling cascades

was evaluated by western blotting. Dopamine-activated D1-D2 heteromer signaling was evaluated by immunocytochemistry in postnatal sex-differentiated male and female primary rat striatal cultures, transduced with adeno associated viral (AAV) vectors expressing the D1-D2 interfering peptide or a control vector. Data were analyzed by two-way-ANOVA (sex and treatment factors), with significance determined by Tukey's or Šídák's post-hoc analysis. Sucrose preference testing was analyzed by two-way repeated measures ANOVA with Tukey's post-hoc analysis.

Results: Results: For both sexes, 24 days of CUS reduced total body weight ($p < 0.0001$), sucrose preference ($p < 0.0001$), open arm elevated plus maze exploration ($p < 0.0001$), and interaction time with a novel conspecific rat ($p = 0.0002$), indicating anhedonia-, anxiety- and social withdrawal-like behaviours ($N = 7-8$ per sex/group). Female rats exhibited shorter latency to reduced sucrose preference at 8 days of stress exposure ($p = 0.022$), indicating enhanced susceptibility to anhedonia-like behaviour. Serum corticosterone was elevated in stress-exposed rats of both sexes on day 7 ($p = 0.003$), but was blunted by day 24. Female rat prefrontal cortex (PFC; $p < 0.0001$), caudate putamen (CPu; $p = 0.003$), nucleus accumbens (NAc) shell ($p < 0.0001$), NAc core ($p < 0.0001$) and amygdala ($p = 0.0004$) exhibited greater basal D1-D2 heteromer density compared to males ($N = 4-5$ per sex/group). CUS induced increased D1-D2 heteromer expression in female PFC ($p = 0.016$) with no effect in males. In NAc shell, CUS downregulated the basally lower D1-D2 heteromer levels in males ($p = 0.022$) while females maintained elevated basal levels, suggesting an aversion-compensating mechanism present in males but absent in females. CUS triggered elevated phosphorylation of CaMKII α ($p = 0.001$), BDNF levels ($p = 0.02$) and tropomyosin receptor kinase B activation (TrkB; $p = 0.048$), exclusively in female rat striatum ($N = 4$). In contrast, CUS induced elevated PDYN ($p = 0.046$) and κ -opioid receptor (KOR; $p = 0.0049$) expression exclusively in male rat striatum ($N = 4-5$). The D1-D2 heteromer-interfering peptide reversed the ability of dopamine to increase BDNF ($p = 0.0051$) and PDYN ($p = 0.0005$) production in primary striatal neurons of both sexes ($N = 3-4$). In rats of both sexes, acute interfering peptide administration reversed the effects of CUS in precipitating anhedonia ($p = 0.0001$) and anxiety-like ($p = 0.0002$) behaviours, without affecting locomotor behaviour ($N = 7-8$ per sex/treatment).

Conclusions: These results highlight the critical role of the D1-D2 heteromer in mediating basal sex differences, which give rise to divergent male- and female-specific mechanisms of stress vulnerability. D1-D2 heteromer activation induces the core behavioral features of MDD symptomatology and contributes to the heightened susceptibility of females to these behaviours, while its inhibition offers a promising therapeutic strategy for MDD symptom reversal.

Keywords: Depression, sex differences, D1-D2 dopamine receptor heteromer, Chronic unpredictable mild stress, Striatal Dopamine signaling

Disclosure: Nothing to disclose.

P271. PERSISTENT CHANGES IN SEROTONIN TRANSPORTER EXPRESSION IN THE HIPPOCAMPUS AND PREFRONTAL CORTEX OF ADULT MALE C57BL/6 MICE FOLLOWING ADOLESCENT FLUOXETINE EXPOSURE

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Background: Prescription rates of fluoxetine, the most dispensed selective serotonin reuptake inhibitor, have been rising among the juvenile population. Since adolescence is a critical developmental period in which the brain undergoes fine-tuning, chronic increases

in serotonin levels may lead to long-term neurobiological changes in brain regions responsible for emotional regulation. Thus, using mice as a model system, we explored how adolescent fluoxetine exposure (AFE) would influence the expression of serotonin transporters (SERT) in the adult hippocampus and prefrontal cortex.

Methods: Postnatal day (PD)-35 male C57BL/6 mice were administered with fluoxetine (250 mg/l in their drinking water) for 15 consecutive days (PD35-49). Three weeks later, when mice reached adulthood (PD70), bilateral hippocampus and prefrontal cortex were dissected and analyzed for SERT protein expression ($n = 10$ per group).

Results: We found that, when compared to water-pretreated controls, AFE-mice exhibited increased SERT protein expression in the hippocampus. In contrast, expression of SERTs was decreased in the prefrontal cortex of these mice.

Conclusions: Our data suggest that AFE induces long-term region-specific changes in SERT expression. Collectively, this study demonstrates that persistent fluoxetine-induced neurobiological changes are evident in adulthood when exposed to this antidepressant medication during the adolescent stage of development.

Keywords: fluoxetine, Antidepressant, SSRI, Hippocampus, dorsolateral prefrontal cortex

Disclosure: Nothing to disclose.

P272. SELECTIVE ACTIVATION OF MOOD-RELATED CIRCUITS WITH LIGHT

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Background: Depression in humans or outcomes of repeated stress in rodents significantly diminishes activity of prefrontocortical circuits and the nucleus accumbens. Previous data has shown that these cortical inputs to Nucleus Accumbens (NAc) dopamine 1 receptor (D1) medium spiny neurons (MSNs) are significantly diminished following repeated stress, which correlates with anhedonic outcomes. We have serendipitously discovered that light flashes can selectively activate this population of cells.

Methods: Light flash activity was monitored using fiber photometry and cell-type selective calcium imaging (NAc GCaMP8f) in Tac1-Cre or A2a-Cre animals which allows for recording activity from NAc core D1-MSNs or D2-MSNs, respectively. In order to test what drives this light flash activity, we used a pharmacological approach where animals were injected (i.p.) with antagonists before eliciting light flash activity. Male and female mice ($N = 4-8$) were used in the study. To evoke plasticity, prefrontocortical (PFC) inputs were injected with the red-shifted opsin chrimson and optogenetic inputs were stimulated in the NAc core through the photometry fiber.

Results: Light flashes evoked D1-MSN calcium activity that was dependent on extrasynaptic ($p < 0.01$) but not synaptic NMDA receptor activity and partially required dopamine 1 receptor activation for sustained and consistent activity. Light flashes evoked multi-phasic dopamine release patterns in the region as measured by dLight fiber photometry. Utilizing protocols for spike timing dependent plasticity, it was found that pairing light evoked D1-MSN transients with prefrontocortical stimulation produced a long term enhancement in PFC evoked input on NAc core D1-MSNs ($p < 0.05$).

Conclusions: Our results indicate that light flashes can assist in the induction of cell type and pathway selective activation of circuits involved in depression. This work may provide a means of

enhancing the rate and efficacy of some depression therapeutics including transcranial magnetic stimulation for treatment resistant depression.

Keywords: Depression, Chronic social stress, Nucleus Accumbens, Plasticity

Disclosure: Nothing to disclose.

P273. SPATIAL LIPIDOMIC AND NEURON-SPECIFIC TRANSCRIPTOMIC SIGNATURES IN THE NUCLEUS ACCUMBENS REVEAL PHOSPHOLIPID DYSHOMEOSTASIS IN STRESS SUSCEPTIBILITY

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Background: Lipid systems play a substantial role in the pathophysiological mechanisms underlying several brain disorders. However, the mechanisms underlying the lipid profile in stress-induced maladaptations and how these changes can impact distinct neuronal circuits, and behavioral states remain to be fully elucidated. Here we studied the effects of gene-environment interaction by using depression-like p11KO mice and chronic stress.

Methods: These mouse models of both sexes were comprehensively analyzed to uncover behavioral, lipidomic, and transcriptomic signatures. In particular, we performed spatial lipidomic analysis via matrix-assisted laser desorption/ionization Fourier-transform ion cyclotron resonance mass spectrometry imaging (MALDI-FTICR-MSI) in dual polarity mode [27] to spatially characterize global lipidome changes in the nucleus accumbens in these mice. Furthermore, we also employed NanoString GeoMx® Digital Spatial Profiler (DSP) technology and fluorescent in situ hybridization on mouse nucleus accumbens tissue to evaluate global and targeted transcriptomic signatures and their association with observed lipid alterations.

Results: Our results show that p11 deficiency and stress interaction enhance susceptibility to depression-like behaviors and provide novel evidence that these responses are associated with significant phospholipid dyshomeostasis in the nucleus accumbens. Phospholipid disturbances were predominantly related to phosphatidylethanolamine (PE) and ether phosphatidylethanolamine metabolism, along with targets involved in the PE biosynthetic pathway. Moreover, chelerythrine administration, a compound reported to disrupt phospholipid balance, significantly induces PE changes and depression-like behaviors.

Conclusions: The present study provides evidence that alterations in phospholipid-related pathways may alter reward/anti-reward circuits and how these changes might be implicated in stress-related disorders.

Keywords: neurobiology, stress, depression, anxiety, s100a10, Lipid metabolism

Disclosure: Nothing to disclose.

P274. CNT-9982, AN OREXIN RECEPTOR 2 AGONIST, ENHANCES WAKEFULNESS IN MARMOSETS, AND IN THE WISTAR KYOTO RAT MODEL OF MAJOR DEPRESSIVE DISORDER, NORMALIZES THE AROUSAL STATE PHENOTYPE AND ALLEVIATES BEHAVIORAL DESPAIR

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Background: Orexin receptor 2 (OX2R) agonists have been shown to enhance wakefulness in rodents, in nonhuman primates, and in clinical studies. Based on an emerging understanding of orexin physiology, it is hypothesized that OX2R agonists may also modulate cognition, mood, and other neuropsychiatric and neurological functions. Low levels of orexin-A (OXA) in cerebrospinal fluid have been shown to negatively correlate with the severity of hypersomnia and mood symptoms in major depressive disorder (MDD). Wistar Kyoto (WKY) rats have reduced numbers of orexin neurons and recapitulate the hypothalamic-pituitary-adrenal axis dysregulation and behavioral symptoms seen in MDD. Here, CNT-9982, an orally available, brain penetrant, highly potent and selective OX2R agonist has been used to interrogate neurobehavior in the WKY rat and healthy non-human primates.

Methods: Ex vivo whole cell current-clamp recordings were performed on brain slices from the ventral tuberomammillary nucleus (TMN) in mouse hypothalamus in the presence of 1 μ M tetrodotoxin, with 3–7 neurons recorded per test concentration of CNT-9982 or OXA. Concentration-response curves were compared using the extra sum-of-squares F-test. In vivo efficacy of oral CNT-9982 on wakefulness, rapid-eye-movement (REM), and non-REM (NREM) sleep was measured during the dark period in 5 male and 3 female marmosets (0.03–1 mg/kg) and in 10 male WKY rats (1–30 mg/kg) in a counterbalanced, repeated-measures design. Undisturbed baseline recordings from WKY vs wild type (WT) wistar rats (each n = 8) were used to determine the arousal state phenotype. Antidepressant-like effects on behavioral despair in WKY rats were evaluated by time spent immobile in the forced swim test (FST) in a between-groups design (n = 10/group). General effects on motor activity were assessed by beam breaks in the open field apparatus in the same WKY rats, and by implanted telemeters, along with core body temperature recordings, in different WKY rats and marmosets. Differences after CNT-9982 vs. vehicle were detected by Holm-Sidak corrected contrasts following analysis of variance.

Results: CNT-9982 depolarized histaminergic neurons from ex vivo mouse TMN brain slices and was significantly more potent than the endogenous ligand, orexin A (EC₅₀ = 7.3 nM vs 31.7 nM, respectively; P < 0.0001). In healthy marmosets, oral CNT-9982 increased EEG spectral power within gamma and alpha bands during wakefulness in a dose and time related manner. In WKY rats, oral CNT-9982 normalized the excessive sleep and low motor activity phenotype observed in the first hour of the active phase; time awake increased (F[1.73,17.3] = 20.5, P < 0.0001) and NREM sleep decreased after 10–30 mg/kg while REM sleep was suppressed after $\geq$ 1mg/kg. The phenotype of decreased EEG spectral power during wakefulness appeared to attenuate within the gamma band after CNT-9982. A single oral dose of CNT-9982 (30 mg/kg) decreased immobility in the FST (F[2,28] = 5.92, P = 0.007, effect size 1.3) without a general effect on motor activity or core body temperature, supporting an acute effect of CNT-9982 in alleviating behavioral despair in WKY rats.

Conclusions: These results support further exploration of OX2R agonists as potential treatments for excessive daytime sleepiness and improvement of mood symptoms in neurological and psychiatric disorders.

Keywords: orexin receptor 2 agonist, orexin/hypocretin, Major depressive disorder (MDD), Wistar Kyoto rat, marmoset

Disclosure: Centessa Pharmaceuticals, Employee, Self.

P275. RESILIENT SPECIFIC SEX-CONSERVED TRANSCRIPTOMIC NETWORKS IN THE NUCLEUS ACCUMBENS FOLLOWING CHRONIC SOCIAL DEFEAT STRESS IN MICE

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Background: Major depressive disorder (MDD) is a leading cause of disability and a major contributor to suicide worldwide. Chronic stress is a key risk factor for MDD and is often modeled in rodents using the chronic social defeat stress (CSDS) paradigm. This model classifies animals into two groups based on their responses to stress: those with depression-like behaviors (susceptible) and those who maintain normal behavior (resilient). However, the CSDS model has been predominantly studied in male mice, leaving a critical gap in understanding female-specific mechanisms, despite MDD being more prevalent in women.

Methods: Identifying sex-specific molecular drivers of stress resilience is essential for advancing personalized treatments. We adapted the CSDS model for female mice and employed RNA-sequencing to analyze transcriptional changes associated with susceptibility and resilience across multiple brain regions. Comparative analyses between sexes were conducted on the nucleus accumbens (NAc), a key brain region involved in stress responses. Weighted Gene Co-Expression Network Analysis (WGCNA) identified gene modules associated with resilience in males and females.

Results: Our findings revealed significant sexual dimorphism in molecular responses to stress. However, approximately 40% of genes upregulated in the NAc of resilient mice overlapped between sexes. WGCNA identified convergent gene modules with one pair showing a 25% overlap—the highest across sexes. Within these modules, GPRIN1 and STX1A emerged as key driver genes positively correlated with resilience. Overexpression of these genes in male mice using viral manipulation induced pro-resilient effects.

Conclusions: This study advances our understanding of sex-specific resilience mechanisms, highlighting molecular pathways that could guide targeted antidepressant development.

Keywords: Depression, stress resilience, sex differences

Disclosure: Nothing to disclose.

P276. LINC00473 REGULATES MITOCHONDRIAL FUNCTION TO PROMOTE FEMALE-SPECIFIC STRESS RESILIENCE

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Background: Depression is a common mental health disorder that causes tremendous personal suffering and places a significant burden on society. Women are twice as likely to suffer from depression as men, yet our understanding of the molecular mechanisms leading to this sex difference is limited. We discovered that long non-coding RNAs (lncRNAs), a prominent class of epigenetic regulators, are robustly and sex specifically regulated in the post-mortem brains of individuals with depression compared to healthy controls. One of the targets we identified is the neuronal gene LINC00473, which is down-regulated in the medial prefrontal cortex (mPFC) of women with depression but not men. Using animal models and human cell culture, we concluded that LINC00473 acts solely in females to drive stress resilience through an unknown mechanism.

Methods: Here, we undertake a mechanistic analysis by performing a comprehensive identification of RNA-binding

proteins using mass spectrometry (ChIRP-MS) for LINC00473 in human female neuroblastoma cells at baseline and after induction of its expression with forskolin (n = 4 per group). In the same setup, we probed the subcellular localization of LINC00473 using RNAscope and its effects on mitochondrial function using Seahorse. We examined the outcomes of expressing LINC00473 in mPFC on female mice's metabolic function in vivo, at baseline and following chronic stress exposure (n = 13 per group). Finally, we generated viral tools to overexpress in neurons one of the ChIRP-MS hits, Aars2 (Aars2-OE), or GFP (GFP-OE) as a control in mPFC of female mice. We assessed behavior in stress-related tasks, including operant lever pressing for liquid reward (n = 12 per group). In addition, we probed mitochondrial gene expression after in Aars2-OE compared to GFP-OE using Magnetic Cell Sorting (MACS) followed by RNA-sequencing (n = 4 per group) or MS (n = 5 per group).

Results: The bioinformatic analysis of ChIRP-MS results revealed that the induction of LINC00473 expression is associated with its binding to mitochondrial proteins. This effect was validated using in situ hybridization for LINC00473 in neuronal cells that showed an increase in LINC00473 expression levels and mitochondrial colocalization after forskolin treatment. Moreover, overexpression of LINC00473 in cultured neurons altered their bioenergetic state and mitochondrial respiration. Additionally, expression of LINC00473 in female mice's mPFC altered metabolic function at the organismic level, as measured by indirect calorimetry. One of the proteins that interacts with LINC00473 and is associated with mitochondrial function is alanyl-tRNA synthetase 2 (Aars2), the enzyme responsible for loading the mitochondrial alanine tRNA. Like LINC00473, Aars2 is downregulated in the mPFC of post-mortem depression cases only in females. To test the causal role of Aars2 in the mPFC of female mice, we compared Aars2-OE to GFP-OE to find a reduction in movement in open field test and a unique behavioral strategy in lever pressing for liquid reward in operant boxes: Aars2-OE mice sampled the non-active lever and magazine in higher frequency than GFP-OE mice while collecting the same amount of rewards. Finally, we profiled Aars2-OE on mitochondrial gene expression compared to nuclear gene expression at both RNA and protein levels to describe the molecular pathways that potentially mediate the effects of LINC00473 and Aars2 on stress-related behavior of female mice.

Conclusions: Our research suggests that changes in mitochondrial function mediate the female pro-resilient effects of LINC00473, and that Aars2 facilitates some of those behavioral and molecular effects. The results shed light on the female-specific mechanisms associated with stress resilience and promote our understanding of sex differences in depression.

Keywords: Depression, Sex differences, Long-non-coding RNA, Mitochondria, mPFC

Disclosure: Nothing to disclose.

P277. EPIGENETIC CROSSTALK BETWEEN H3K27ME1 AND H3K36ME3 IN THE NUCLEUS ACCUMBENS SHAPES SUSCEPTIBLE VERSUS RESILIENT TRAJECTORIES TO CHRONIC STRESS

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Background: Histone post-translational modifications (PTMs) play a crucial role in mediating lasting changes in gene expression induced by chronic stress and can influence resilience or susceptibility to stress-related disorders. By combining unbiased

proteomic profiling and genome-wide approaches, our group recently uncovered a role for mono-methylation of lysine 27 at histone H3 (H3K27me1) in the enduring effects of stress. We found that mice susceptible to chronic social defeat stress (CSDS) displayed increased H3K27me1 enrichment and concomitant depletion of H3K27me2 in the nucleus accumbens (NAc), a key brain region involved in reward processing and mood regulation. We also observed that resilient mice to CSDS exhibited a higher abundance of H3K36me3. Here, we profiled genome-wide accumulation of H3K36me3 in the NAc of mice exposed to CSDS and assessed the potential histone crosstalk between H3K36me3, H3K27me1, and H3K27me2 in shaping susceptible versus resilient trajectories to chronic stress.

Methods: We exposed adult male mice to CSDS, a validated model for studying depression-like behaviors that separates mice into susceptible and resilient populations based on a social interaction test (SIT). NAc tissue from control, susceptible, and resilient mice ($n = 16$ per group, pooled into 4 biological samples per group) was collected 24 h after the SIT and processed for H3K36me3 genome-wide enrichment using Cleavage Under Targets and Release Using Nuclease (CUT and RUN), followed by sequencing.

Results: Our results show that CSDS exposure differentially altered genome-wide H3K36me3 enrichment in the NAc. Specifically, we found that while H3K36me3 was enriched across intergenic regions and depleted across promoter sites in susceptible mice, there was a higher accumulation of H3K36me3 at promoter regions and depletion across intergenic regions in resilient mice to CSDS. Interestingly, we also observed that H3K36me3 enrichment, along with concomitant H3K27me1 depletion, within genes involved in transcriptional regulation, was a defining hallmark of stress resilience. Finally, we are currently utilizing advanced bioinformatics to identify changes in chromatin accessibility and functional regulatory elements that coincide with enrichment of H3K36me3, H3K27me1, and H3K27me2, and that may uniquely drive resilience to stress.

Conclusions: Together, our results suggest an epigenetic crosstalk between H3K36me3, H3K27me1, and H3K27me2 in fine-tuning susceptibility versus resilience to stress, challenging the traditional one-mark-one-function model by demonstrating that histone PTMs act as combinatorial signals in response to stress.

Keywords: Epigenetics, Chronic social stress, Depression, Histone methylation, Epitranscriptomics

Disclosure: Nothing to disclose.

P278. ASSESSING THE IMPACT OF AMBIENT TEMPERATURE ON DAY-TO-DAY COGNITIVE PERFORMANCE IN OLDER ADULTS: A PILOT STUDY EXPLORING WITHIN-AND BETWEEN-PERSON ASSOCIATIONS

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Background: Extreme heat exposure can adversely impact cognitive function and increase the risk of Alzheimer's disease and related dementias (ADRD). As global temperatures continue to rise, there is concern that heat may accelerate neurodegeneration, yet heat remains unrecognized as a modifiable risk factor, and limited research exists on the causal relationships between extreme heat and cognitive decline. This pilot study aimed to examine both the between- and within-person associations between heat and real-world cognitive performance among older adults.

Methods: Nine participants, aged 70–92 (mean age = 78, $SD = 8$) living in College Station, TX, completed a suite of nine NeuroUX cognitive ecological momentary assessments on their smartphones daily for 14 days in September of 2024. These assessments measured recognition memory, working memory, processing speed, and executive functions. Temperature data was collected via GENEActiv accelerometer throughout the 14-day data collection period. A random-intercept multilevel model was used to examine the dynamic relationship between temperature and daily cognitive performance. To distinguish within-person from between-person effects, the model included both momentary temperature (centered within-person) and the average temperature for each participant across the assessment period (between-person effect). Study day was also included in all models to account for practice effects. Partial R^2 values for fixed effects were estimated using the Nakagawa-Schielzeth-Johnson method.

Results: Adherence to the mobile cognitive testing was excellent, ranging from 86 to 100% (mean adherence = 93%). The average mean temperature during data collection was 86.0°F ($SD = 1.3^\circ F$; range = 84.2°F – 88.3°F), and the average standard deviation in temperature was 3.7°F ($SD = 0.7^\circ F$). Although not statistically significant due to the small sample size, small to medium effects were observed in the relationships between average cognitive test performance and average temperature: working memory (Partial R^2 s: Memory Matrix 0.088; Matching Pair = 0.132; Odd One Out = 0.044), executive functions (Partial R^2 s: Color Namer = 0.121; HandSwype = 0.091) and recognition memory (Memory List Partial $R^2 = 0.097$).

Conclusions: Results of this pilot study did not find clear evidence that cognition fluctuated with temperature within individuals over the 14-day period. This likely reflects the limited variation in temperature during the study (average SD of just 3.7°F) and that the temperature remained warm but not extremely hot (average 86.0°F), which constrained our ability to detect within-person effects. Although none of the between-person effects reached statistical significance, several showed small to medium effect sizes accounting for 4.4% to 13.2% of the variance in cognitive functioning. This suggests that higher overall average temperatures across days may be associated with slightly worse cognitive performance, characterized by lower scores and longer reaction times. Future research with greater temperature variability and larger samples is needed to better understand these relationships to inform public health strategies to mitigate heat-related cognitive decline among vulnerable older adults.

Keywords: Ambulatory Assessment, Alzheimer's Disease, Unsupervised Cognitive Testing, Environmental exposures, Modifiable Risk Factors

Disclosure: Nothing to disclose.

P279. PREDICTION OF DEPRESSION AND HYPO/MANIA IN YOUNG PEOPLE WITH BIPOLAR DISORDER: CONTRIBUTIONS OF MOBILE SENSING METRICS

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Background: Given that bipolar disorder (BD) is a recurrent illness, improving the imminent prediction of mood recurrences could facilitate more timely intervention and improve outcomes. Mobile sensing data, continuously and passively collected from smartphones, may identify behavioral changes that predict mood recurrence. Here, we assess whether mobile sensing metrics, particularly those that map onto constructs critical to BD

recurrence (e.g., sleep and activity), predict next-week depression and hypo/mania in young people with BD.

Methods: In this ongoing study, we recruited young people (14–25 years old) with BD-I/II, currently in remission, and assessed them every 6 months (up to 24 months). Weekly mood status was assessed via the Longitudinal Interval Follow-up Evaluation – Psychiatric Status Rating (PSR). Using the AWARE app, mobile sensing data, including activity classification, screen unlocks, and GPS were collected without requiring user input. As a proxy for Total Sleep Time, we calculated the Total Continuous Offline Time (TCOT), defined as the longest period overnight with minimal activity or smartphone unlocks. We used linear mixed models to test which mobile sensing streams predicted next-week mood episodes; all analyses were adjusted for age and sex, and corrected for multiple comparisons.

Results: Thus far, 35 participants have contributed a median of 33 weeks of sensor data and PSR mood ratings. Depression was predicted by less physical activity (duration mobile: OR = 0.68, $p = 0.0001$) and more smartphone usage (duration screen unlocks: OR = 1.44; $p = 0.0001$) the previous week. Hypo/mania was predicted by more physical activity (OR = 1.25, $p = 0.01$), less time stationary (OR = 0.61, $p < 0.0001$), and more variable sleep (TCOT SD; OR = 1.34, $p = 0.007$) the previous week. All within-person models were also significant, indicating that within-person changes in mobile sensing metrics predicted next-week mood state.

Conclusions: Preliminary analyses from our ongoing longitudinal study of youth with BD indicate that mobile sensing metrics may predict next-week mood state. Limitations include small and homogeneous sample (89% female, 74% white). Future analyses will assess the degree to which models incorporating mobile sensing data provide accurate, person-level prediction, which would pave the way for just-in-time assessments and intervention.

Keywords: Bipolar Disorder, mobile sensing, youth and young adults

Disclosure: Nothing to disclose.

P280. OBJECTIVELY MEASURED SMARTPHONE USAGE IN YOUTH IS ASSOCIATED WITH CHANGES IN LONELINESS AND PSYCHOLOGICAL WELL-BEING

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Background: There has been significant controversy over the association between technology use and youth mental health outcomes, which has led to widespread debate on the best approaches to manage device usage in children and adolescents in order to minimize adverse outcomes. While many studies have reported that social media use in particular is associated with poor mental health functioning, these findings are not universally supported. Recognized limitations of existing work include high reliance on self-reported device usage – which has been shown to be unreliable; and overly coarse characterization of device usage (e.g., total screen time/day). The current analysis sought to use objectively measured indicators of specific types of technology use (ie., social media versus gaming) and also examined how the key demographic variables of age and sex-at-birth interact with these device usage variables to predict changes in self-reported loneliness and overall psychological well-being across a 1-month time frame.

Methods: Data for this study were derived from the Technology Exposure and Child Health: Social Impact and Social Effects (TECHWISE) study, an ongoing, national prospective study

(NCT06664944) examining objective device usage patterns measured through a commercially-available smartphone app and a range of social, psychiatric, and mental health outcomes in children and adolescents aged 8–17 years. For this analysis, total screen time and the percentage of daily screen time by social media and gaming apps were averaged between baseline and month 1 for users with at least five days of usage data. An ElasticNet 10-fold cross-validation modeling approach, which merges Ridge and LASSO regression methods, was used to predict 1-month change in loneliness as measured by the UCLA Loneliness Scale, Revised; and overall psychological well-being, as measured by the 5-item World Health Organization Well Being Index (WHO-5). The model included 13 input features: baseline self-report total score, average daily device use, percentage of time in app categories (social or gaming), binarized age (>12 years) and sex at birth, and their interaction terms. Model performance was evaluated using Mean Squared Error (MSE) and R², with non-zero coefficients in the final model considered statistically significant.

Results: The analysis included data from 204 participants aged 8 to 17 years (mean age = 12.25; 102 female; 111 aged 12 years or younger). The ElasticNet model demonstrated improved predictive performance over a baseline model that predicted the average change in loneliness scores (Model MSE = 41.78 vs. Baseline MSE = 49.05; Model R² = 0.148) and well-being (Model MSE = 250.61 vs. Baseline MSE = 325.06; Model R² = 0.229). This confirms that screen usage features provided valuable predictive information beyond the baseline score. The most important predictor of change was the baseline score; however the model also identified several significant predictors of changes in loneliness and well-being. A higher percentage of total screen time dedicated to social media was associated with increased loneliness over one month and for those over 12 social media use was associated with decreased well-being. Total device usage was not predictive of changes in either outcome. Percentage of time gaming was associated with increased well-being. Furthermore, a significant interaction between age and sex indicated that teenage females are more likely to experience an increase in loneliness and decreased well-being compared to males and younger girls.

Conclusions: We found that in a representative sample of US children and adolescents, an increased proportion of device usage devoted to social media significantly predicted increases in self-reported loneliness over a 1 month period, while proportion of device time spent gaming predicted an increase in overall well-being. We also found that, independent of device usage, older females report more increases in loneliness. These findings extend previous work by using objective (versus self-reported) indices of device usage, and emphasize the importance of a nuanced characterization of device usage in relation to mental health outcomes. Total daily device usage, which has often been used as a potential correlate of mental well-being outcomes, is not associated with changes in loneliness or well-being, whereas social media and gaming specific use as a proportion of overall use are. Current findings are not without limitations. First, we broadly characterized app categories and did not look at the potential relative contributions of specific social media or gaming apps (e.g., TikTok, Roblox). Second, we looked at all device usage variables as an overall average across a month. To fully understand potential causal effects, it will be important to understand potential temporal trends in how device usage changes over time are associated with outcomes over the same time frame. These findings and the broader study from which they were derived are an important step toward a more comprehensive and actionable understanding of how device usage impacts youth outcomes.

Keywords: Social Media Use, Children and Adolescents, smartphone apps, loneliness

Disclosure: Aura, Employee, Self.

P281. CAN AI REPLACE ME? AN EXPLORATORY STUDY OF ARTIFICIAL INTELLIGENCE PERFORMANCE ACROSS ACADEMIC PHYSICIAN TASKS

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Background: Artificial intelligence (AI) platforms are increasingly integrated into medical and academic workflows, yet systematic evaluations of their ability to approximate physician-scientist tasks are limited. Given the hype and fears surrounding AI adoption, we sought to assess current capabilities of multiple AI systems across the three central responsibilities of an academic physician: research, teaching, and clinical care.

Methods: Three representative tasks were selected for each domain and results were compared to a previously prepared human reference or assessed by blinded independent rater review. Three AI platforms were tested (ChatGPT, Perplexity, Open Evidence). Research tasks included: (1) Editing of abstracts for conference submission, evaluated by a blinded independent rater; (2) generation of a review article outline compared against an expert human reference; (3) identification of NIH reviewer criticisms from prior grant submissions. Teaching tasks included: (1) Identification of criticisms and future directions from a research article compared against prior journal club presentations; (2) identification of key material for a clinical lecture; (3) generation of appropriate test questions from lecture material. Clinical tasks included: (1) Clinical solution for medication management cases compared to solution obtained by prior extensive literature review; (2) development of behavior therapy plans for three cases, rated independently; (3) accurate completion of administrative tasks (school letters, rating scale scoring/feedback, progress notes).

Results: AI performance varied by domain and platform. In the research domain, AI produced adequate criticisms, edits and outlines, but lacked human nuance. Across the teaching domain, performance was strong. In the clinical domain, AI treatment planning was expert level and medication options included the physician's chosen intervention; however, administrative clinical tasks frequently triggered hallucinations and inaccurate outputs.

Conclusions: AI already demonstrates utility in supporting academic physician workflows, particularly in teaching, elements of research and treatment planning. However, oversight remains essential, as nuanced judgment and complex administrative tasks remain challenging. Given the rapid pace of model development and decreasing error rates, AI is poised to become an important adjunct, not a replacement, to physician-scientist performance.

Keywords: Generative Artificial Intelligence, Career Development, academic performance

Disclosure: myriad genetics, Advisory Board, Self, Tourette Association of America, Advisory Board, Self.

P282. HERITABILITY OF ACCELEROMETRY-DERIVED FEATURES OF PHYSICAL ACTIVITY, SLEEP AND CIRCADIAN RHYTHMICITY AND THEIR JOINT INFLUENCES IN YOUNG PEOPLE

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Background: Dysregulation of motor activity and sleep are key components of several mental disorders, particularly mood

disorders and attention deficit hyperactivity disorder (ADHD). There is growing evidence from studies that employ accelerometry that profiles of 24-h patterns of activity may discriminate subgroups of mood disorders and ADHD in adults. However, there is limited information on activity profiles in youth that may identify differential risk for psychopathology across development. The aims of this paper are: (1) to examine the familial and genetic influences on 24 h patterns of objectively-assessed activity in young people based on genetically informative samples; and (2) to identify age, sex, and contextual influences on heritability; and (3) to evaluate whether PolygenicScores (PRS) for sleep, activity, mood, and attention are related to empirically derived estimates of these domains.

Methods: Samples include three studies of youth from genetically informative study designs including families and twins. Family study data included youth ages 10–25 from the National Institute of Mental Health (NIMH) Family Study of Affective Spectrum (n = 154) and the Lausanne-Geneva Family Study (PsyCoLaus, N = 176). Twin study data included 495 youth ages 9–17 from the Brisbane Adolescent Twin Study (BATS) and the Queensland Twin Adolescent Brain (QTAB) project (N = 93 monozygotic; 117 dizygotic (DZ) twin pairs, and 75 unmatched twins). Accelerometry was assessed with the GeneActiv device for two weeks as part of the Motor Activity Research Consortium for Health (mMARCH) collaborative study. Polygenic heritability estimates (the proportion of the total phenotypic variance explained by additive genetic (or familial) effects) adjusted for age and sex were derived from the Sequential Oligogenic Linkage Analysis Routines (SOLAR) software package for the features of the three domains of physical activity, sleep, and circadian rhythmicity. Heritability of the latent structure of the individual and joint factors was estimated with an integrative data reduction technique for multi-modal multivariate data, termed "Joint and Individual Variation Explained" (JIVE). Associations between heritable domains with polygenic scores (PRS) for a range of related domains were also examined.

Results: Heritability estimates of specific features of sleep, activity, and circadian rhythmicity range from 0.44 to 0.87 and were highly significant in both the twin and family studies, with an increase in heritability with increasing age. Heritability estimates of the variability of many salient features was >averages (0.56–0.89), suggesting that heritability of the dynamics of these domains may supersede that of the actual levels of sleep and activity. Strong environmental influences suggested by weekday-weekend differences also highlight the importance of contextual factors in examining these systems. Finally, the heritability of the JIVE estimates of joint factors was lower than that of the individual factors.

Conclusions: The increase in the influence of genetic factors for sleep, physical activity, and circadian rhythms across adolescent development has important implications for prevention and intervention. The lower heritability of the joint cross-domain factor than those of the individual domains suggests that there may be distinct genetic and biologic factors underlying these systems. The associations with PRS for some of these domains highlights the promise of objective monitoring of these systems in young people that could facilitate prevention and intervention of disorders of sleep, mood and activity.

Keywords: Sleep, circadian rhythms, locomotor activity, twins, youth and young adults

Disclosure: Nothing to disclose.

P283. GENETIC ANALYSIS LINKS RODENT LOCOMOTOR ACTIVITY TO HUMAN EXTERNALIZING BEHAVIOR

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Background: The locomotor response to a novel environment is widely studied in mice and rats. This trait is hypothesized to be related to human externalizing behaviors and substance use disorders. However, there are limited empirical data to support this hypothesis.

Methods: We used data from a genome wide association study (GWAS) in outbred rats ($n \sim 8000$) and a large human GWAS of human externalizing behavior (effective $n \sim 1,500,000$) to empirically evaluate the relationship between these two traits. We used NetColoc to examine the overlap between the networks of genes identified by these two datasets.

Results: We found that the networks interconnecting these two traits showed substantial and highly significant overlap ($p = 2.03 \times 10^{-6}$), despite insignificant overlap between locomotor and externalizing activity at the gene level ($p = 0.51$). The network showed enrichment for neurobehavioral and nervous system genes. We also found significant network overlap between locomotor activity and other SUD-related traits, Parkinson's disease, and ADHD.

Conclusions: Our results provide empirical evidence supporting the role of rodent locomotor activity as a model for human externalizing behavior and SUDs, validating its use to dissect the molecular, genetic, and neurological basis of substance abuse.

Keywords: Externalizing behavior, locomotor activity, GWAS

Disclosure: Nothing to disclose.

P284. THE ROLE OF LET-7 IN TRANSGENERATIONAL TRANSMISSION OF OPIOID-RELATED PHENOTYPES

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Background: The United States is in the midst of an opioid addiction crisis, with exposure rates to opioids estimated to be over 3.3% of the population in 2021 alone. In addition to the devastating consequences to the current population, which include addiction, overdose, lost productivity, socioeconomic burdens, and social and familial deterioration, a growing body of evidence indicates that opioid exposure will also affect subsequent generations and contributes to the inheritance of increased susceptibility to develop opioid use disorder. The current set of experiments uses a rat model to determine the impact of adolescent opioid exposure on the expression of the microRNA let-7g in the testes and spermatozoa, because it is known to regulate the mu opioid receptor. We also determine the impact on future generations by examining mu opioid receptor expression and oxycodone self-administration in the offspring and grandoffspring. Lastly, we use human sperm samples to determine if let-7 is also upregulated in the testes of men with a history of drug experience.

Methods: Male Sprague Dawley rats were administered increasing doses of morphine (5–25 mg/kg, s.c.) for 10 days during adolescence (P30–39). Rats were then maintained drug free until adulthood (P70–80) at which point they were mated with drug-naïve females. After mating, Sal-F0 and Mor-F0 males were euthanized and the testes and spermatozoa collected. Let-7g was measured via qRT-PCR in these tissues. In the offspring (Sal-F1 and

Mor-F1), *oprm1* was measured on PND1, PND30, and PND60 from the ventral tegmental area (VTA) and nucleus accumbens (NAc). In adulthood, one male and one female from each litter were tested for oxycodone self-administration (0.1 mg/kg/infusion, 2 h sessions). A naive male littermate was used to breed F2 grandoffspring, which were tested for oxycodone self-administration in adulthood.

Results: Results demonstrate increased levels of Let-7g in the sperm ($t_{14} = 3.8$; $p < 0.01$) and seminiferous tubules ($t_{15} = 2.4$; $p < 0.05$). The almost 8-fold increase in let-7g expression in sperm compared to 1.5-fold increase in let-7g expression in testes may indicate that let-7g in the testes is located within progenitor cells, spermatogonia, or sperm cells and the signal to noise was decreased due to the presence of additional cell types. Moreover, opioid receptor expression on Sertoli and Leydig cells of the testes indicate potential long-term effects of adolescent opioid exposure on cells involved in spermatogenesis. Results from the PND1 brains revealed a significant increase in *oprm1* in Mor-F1 animals compared with Sal-F1 animals ($p < 0.05$). Protein expression demonstrated increased mu opioid receptor expression in the NAc. Lastly, male and female Mor-F1 animals self-administered more oxycodone than the respective Sal-F1 control groups. These data revealed a significant main effect of sex $F(1, 570) = 39.1$, sire $F(1, 570) = 106.4$, and day $F(14, 570) = 13.9$. For the males, there was a significant main effect of both sire exposure $F(1, 22) = 16.47$ and day $F(14, 308) = 9.847$. For the females, there was a significant main effect of sire exposure $F(1, 16) = 4.591$, day $F(14, 224) = 13.45$, and a significant interaction $F(14, 224) = 2.04$. Preliminary results examining self-administration in F2 animals showed a significant decrease in oxycodone self-administration in Mor-F2 males compared with Sal-F2 males, and no differences in the females.

Conclusions: This set of data suggests that let-7g in sperm is a persistent mark that may regulate mu opioid receptor expression within the testes. Because it was present in such high levels in the sperm of Mor-F0 animals, we hypothesized that let-7g is involved in the transmission of the molecular and behavioral phenotypes observed in Mor-F1 offspring. Indeed, we measured *oprm1* expression from the midbrain of PND1 offspring and saw increased *oprm1* transcript in both males and females supporting our hypothesis that let-7g is disrupting expression of the mu opioid receptor protein in F1 offspring, which may underlie the observed increases in oxycodone self-administration during adulthood. Ongoing studies are using transfection of let-7g mimics and inhibitors in spermatozoa, paired with artificial insemination to rescue or recapitulate the offspring phenotype. Additional experiments are examining if let-7g is a translational target by measuring this miR in men with and without a history of drug exposure.

Keywords: Epigenetics, MicroRNA, Mu opioid receptor, transgenerational transmission, oxycodone

Disclosure: Nothing to disclose.

P285. GENETICALLY MODELED GLP1R AND GIPR AGONISM REDUCE BINGE DRINKING AND ALCOHOL-ASSOCIATED PHENOTYPES: A MULTI-ANCESTRY DRUG-TARGET MENDELIAN RANDOMIZATION STUDY

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Background: Pharmacological modulation of glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide

(GIP) through dual GIP/GLP-1 receptor agonists, commonly used for diabetes and obesity, shows promise in reducing alcohol consumption.

Methods: We applied drug-target Mendelian randomization (MR) using genetic variation at these loci to assess their long-term effects on problematic alcohol use (PAU), binge drinking, alcohol misuse classifications, liver health, and other substance use behaviors.

Results: Genetic proxies for lowered BMI, modeling the appetite-suppressing and weight-reducing effects of variants in both the GIPR and GLP1R loci ("GIPR/GLP1R"), were linked with reduced binge drinking in the primary ($\beta = -0.44$, 95% CI $[-0.72, -0.15]$, $P = 2.42 \times 10^{-3}$) and replication data ($\beta = -0.13$, $[-0.22, -0.04]$, $P = 0.0058$). HbA1c lowering via GIPR/GLP1R variants was associated with reduced risk of heavy drinking with psychiatric comorbidities versus low-risk drinking (odds ratio [OR] = 0.62, [0.45, 0.85], $P = 0.0031$), with replication in independent HbA1c data (OR = 0.71, [0.60, 0.84], $P = 5.22 \times 10^{-5}$) and directional consistency with reduced PAU. Analysis of individual loci indicated that both GIPR and GLP1R were protective against heavy drinking, underscoring the importance of both targets. While estimates for other substance use disorders (tobacco, cannabis, opioid) were consistently null, food preference analyses revealed that BMI lowering via GIPR/GLP1R reduced fatty food liking ($\beta = -1.58$, $[-2.01, -1.14]$, $P = 1.62 \times 10^{-12}$) and increased vegetarian food liking ($\beta = 2.08$, [1.17, 2.99], $P = 8.22 \times 10^{-6}$), implicating metabolic and appetite regulation pathways for the alcohol consumption findings. Consistency across multiple MR methods and colocalization analyses strengthened causal inference.

Conclusions: Mediation analysis suggested reductions in hazardous alcohol consumption partially explain the cardioprotective effects of these agonists. Multi-ancestry analyses supported directionally aligned relationships in non-European cohorts. These findings support further clinical exploration of GLP1R, GIPR, and dual agonists in addiction medicine.

Keywords: Alcohol, GLP1, Human Genetics, Mendelian Randomization, alcohol drinking

Disclosure: Nothing to disclose.

P286. ALCOHOL DEPENDENCE OR INTAKE DOES NOT ALTER BLOOD LEVELS OF THE INFLAMMATORY ADIPOKINE LIPOCALIN-2 IN RATS OR HUMANS

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Background: Inflammation, including low-grade inflammation, plays an important role in alcohol use disorder (AUD) pathophysiology. Yet, there is still much to be understood about the role of inflammation in AUD. One inflammatory peptide whose role has not been thoroughly investigated in relation to AUD is lipocalin-2 (LCN2). Lipocalins are extracellular proteins that are transporters for hydrophobic molecules such as retinol, steroids, pheromones, and siderophores. LCN2 was originally discovered in human neutrophils and is primarily secreted from adipose tissue. Peripherally produced LCN2 crosses the blood-brain barrier. LCN2 is an inflammatory adipokine and is proposed as a biomarker for kidney and heart injury, metabolic disease, and breast cancer. In addition, LCN2 may regulate stress and anxiety-related behaviors and neuroadaptations that occur in response to stress. Although LCN2 expression is typically absent in the brain, it is highly expressed in the central nervous system (CNS) in response to injury and inflammatory stimuli and may modulate

neuroinflammation. Additionally, human and rodent studies show that LCN2 drives alcohol-induced inflammation, propagating the development of alcohol-associated liver disease. Yet, there is a paucity of knowledge about LCN2 in the context of alcohol intake and AUD. To our knowledge, LCN2 levels have not been studied in a model of alcohol dependence.

In this preliminary study, we examined blood levels of LCN2 in the context of alcohol use and dependence in humans and rats. We compared LCN2 levels in alcohol vapor-exposed rats with those of nondependent controls. In humans, we investigated the effect of alcohol cues or alcohol administration on blood LCN2 levels in cue-induced alcohol reactivity or after alcohol administration. We hypothesized that there would be alterations in LCN2 levels in alcohol-dependent rats and humans with heavy alcohol drinking.

Methods: 17 male Wistar rats were used, 9 of which were made dependent on alcohol starting at 10 weeks of age by chronic, intermittent alcohol vapor exposure. This protocol has been previously described and leads to alcohol dependence. These animals were exposed to daily cycles of 14 h of alcohol vapor, followed by 10 h of room air, for 12 weeks prior to blood collections. Blood was collected at 8:00am (end of vapor exposure; intoxication) and 4:00pm (8 h into alcohol withdrawal). Non-dependent rats ($n = 8$) were not exposed to alcohol vapor at any point in time. The vapor model of alcohol dependence has been shown to produce several somatic and affective symptoms of alcohol dependence, including escalated and compulsive-like alcohol consumption. Blood was collected when animals were 22 weeks old. Blood was drawn at 8:00 am and 4:00 pm.

Blood LCN2 levels were also examined in two human laboratory studies with cue-reactivity, oral and intravenous alcohol administration in participants with AUD and a DSM-IV diagnosis of alcohol dependence ($n = 15$ and $n = 16$, respectively). The first human study consisted of male and female patients with AUD, and the second study consisted of heavy alcohol-drinking individuals with a weekly alcohol consumption of >15 and >20 standard drinks for women and men, respectively. The primary goal was to evaluate the effects of different pharmacological interventions on alcohol-related outcomes. For this secondary analysis of LCN2 levels, blood samples from the placebo arms only were analyzed. While the rats were male only, the two clinical studies included both male and female participants.

LCN2 levels were measured by ELLA Protein Simple system. LCN2 levels in rats were analyzed using a mixed-methods model, with dependence (alcohol-dependent vs. non-dependent) as a between-subjects factor and Time (8:00 am vs. 4:00 pm) as a within-subjects factor. LCN-2 levels in humans were analyzed using linear mixed-effects models, with repeated Times as a fixed effect, individual participants as a random effect, and age, sex, race, and body mass index (BMI) as covariates.

Results: Blood measurements indicated a significant effect of Time in rats (i.e., morning versus afternoon; $p = 0.0004$) but not of dependence on LCN2 levels. In humans, no significant effects were found on LCN2 levels, indicating that exposure to alcohol cues or oral/intravenous alcohol administration did not change blood LCN2 levels. Given that we did not have an a priori hypothesis in terms of sex differences, plus the unbalanced number of males and females, we did not test the LCN2 data in humans for sex differences. Nonetheless, sex was included in the human studies as a covariate in the analyses and no significant effects were detected.

Conclusions: We did not find differences in plasma LCN2 levels between non-dependent rats and alcohol-dependent rats during intoxication or withdrawal. Results in individuals with alcohol dependence showed that exposure to alcohol cues or alcohol administration (oral or IV) did not change plasma LCN2 levels. The Time effect seen in male rats was likely related to circadian fluctuations of LCN2 levels, regardless of alcohol exposure or

dependence, with levels being higher in the morning (at the beginning of the active, dark period) than in the afternoon. The present study contributes rodent and human evidence to better understand the neurobiology and correlates of AUD from a mechanistic perspective. Additional studies are needed to better understand the potential link between LCN2, alcohol use, and circadian variations.

Keywords: Inflammation and cytokines, Alcohol dependence, Addiction

Disclosure: Nothing to disclose.

P287. SIMMERING PASSION: PILOT TESTING OF A DOPAMINE D3-PREFERRING PARTIAL AGONIST (CARIPRAZINE) IN A TASK THAT MEASURES BOTH 'FIRST ORDER' AND 'SECOND ORDER' CUE-REACTIVITY IN INDIVIDUALS WITH OPIOID AND COCAINE USE DISORDER

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Background: Our laboratory pursues druggable, relapse-relevant endophenotypes ('brain biomarkers') as potential treatment targets in addiction. One well-characterized endophenotype is the learned response to drug "reminder" cues: over the natural course of drug use, signals for the arrival of the drug come to trigger physiological arousal, drug anticipation/desire ("craving"), and activation of brain networks underlying reward and motivation.

Classically-conditioned cues and responses that develop in the context of actual drug arrival are termed 'first order' conditioned effects. Importantly, we can learn many "second order" associations, linking signals that are remote from the actual biological event of interest. For example, just turning the corner to go down the bakery street can come to trigger salivation and pastry desire; well before experiencing the powerful 'first order' cues, i.e., the sights and smells of the pastries that are closely and more directly linked to eating the croissant.

We reasoned that individuals who show more 'second-order' conditioning might be more vulnerable to cue-triggered relapse. For the current project, we designed a new task, SOC-R (Second-Order Conditioning - with Recognition probes) to reveal potential 'second-order' effects (see Methods).

In addition, as D3 partial agonists/antagonists can block conditioned effects in both pre-clinical (e.g., reinstatement and conditioned place preference) and clinical (ACNP 2020) paradigms, we are testing whether the strongly D3-preferring agent cariprazine can blunt (first-order or second order) cue effects in the SOC-R. Thus, these studies are designed both to offer a window onto second-order conditioning as a useful dimension of the cue-vulnerable endophenotype, and to assess the potential of D3 (partial agonist) agents to impact this vulnerability.

Methods: Participants were treatment-seeking individuals diagnosed with Opioid Use Disorder and a Cocaine Use Disorder ($n = 5$ thusfar), stable on methadone or buprenorphine, who gave witnessed, informed consent. Inpatient stabilization and induction onto cariprazine (1.5 mg daily) vs. placebo across 10–14 days was followed by an event-related fMRI session that included the SOC-R task. The initial ("Conditioning") segment of the SOC-R task featured 48 cocaine-related trials (500 ms turquoise squares immediately preceding a 500ms cocaine image atop the square) and 48 non-drug trials (500 ms purple circle immediately preceding a 500 ms non-drug image of a chair, atop the circle),

in quasi-random order. The second ("Testing") segment of the task featured quasi-random presentation of only the geometric shapes [e.g., turquoise squares or purple circles], along with additional novel and familiar non-drug stimuli. Data were smoothed, normalized, realigned and batch-analyzed within the SPM 12 pipeline. Pre-planned contrasts compared the brain response to cocaine vs. neutral cues in the Conditioning segment, and to turquoise squares vs. purple circles in the "Testing" segment, to assess for second-order effects. The resulting statistical parametric maps were thresholded at $1.65 < t < 5$ for display and examination, including medication status (cariprazine vs. placebo).

Results: In the 'Conditioning' segment of the SOC-R task, the two placebo patients showed drug cue-triggered activation of the brain's motivational circuitry (also including OFC and visual association regions), while three patients receiving 1.5 mg of daily cariprazine showed 'quiet' brain maps. In the 'Testing' segment, there was indeed evidence of differential activation to the turquoise square (visual regions, OFC, striatum and insula) in the two placebo patients. The two cariprazine patients showed some potential blunting of the striatal response to the turquoise square, but the brain response in extra-striatal regions was not completely eliminated ($p < 0.05$ uncorrected for this preliminary examination).

Conclusions: These findings offer preliminary evidence for second-order conditioning of limbic arousal to an arbitrary (geometric shape) stimulus. This metric may complement assessment of the more conventional 'first-order' cue effects, and may have potential clinical significance: in our prior studies, individuals with heightened second-order conditioning of arousal to neutral cues (in a cocaine cue-task) were at increased relapse risk.

To our knowledge, SOC-R is the first task to offer quantification of both the first-order and second-order cue effects. This distinction may be useful, as the research literature suggests that once established, second-order conditioned effects are highly resistant to extinction, even when the first-order effects are extinguished or devalued. Second-order learning may thus represent a passion that is long-lived and insidious, simmering silently unless revealed by a second-order task design.

Though cariprazine at 3 mg was effective in blunting the first-order conditioned drug cue effects (in our prior cocaine population), in the current study using 1.5 mg, some second-order effects were evident despite the medication. Further studies at both doses will be needed to determine the role of dose vs. the type (first- or second-order) conditioned effects. Finally, complementing pharmacologic approaches, neuromodulatory interventions that can reach deep limbic regions (e.g., Low Intensity Focused Ultrasound) may offer a very direct approach for targeting the brain circuitry underlying the drug cue endophenotype.

Keywords: second order conditioning, drug cue reactivity, cariprazine

Disclosure: Nothing to disclose.

P288. SEX AND FAMILY HISTORY OF ALCOHOL PROBLEMS AS MODERATORS OF ALCOHOL ANALGESIA

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Background: Self-management of pain using alcohol is common and is associated with greater risk for chronic pain and alcohol-related problems, including alcohol use disorder. Laboratory-based and observational studies suggest acute alcohol intake has dose-dependent analgesic effects. Furthermore, a substantial

literature supports the notion of sex differences in alcohol-related risk and pain prevalence and severity. At the same time, family history of alcohol problems (FH) is known to influence subjective alcohol response and risk for alcohol-related problems. However, it is not well understood whether sex and FH act as moderators of alcohol analgesia. In this analysis, we combined data from two acute alcohol challenge studies to address this gap. We hypothesized that alcohol's analgesic effects would be stronger in FH+ than FH- individuals. Based on pilot studies and opioid-related data, we also hypothesized that women would experience greater reduction in pain sensitivity than men.

Methods: Data from two NIH-funded studies with highly similar designs were combined for this secondary analysis (Study 1: R01AA025337, Study 2: R21AA026805, JB PI). $n = 112$ participated in Study 1 and $n = 51$ participated in Study 2, for a total $N = 163$ (53.4% women; 36.8% FH+). In both studies, participants completed two counterbalanced, double-blind laboratory sessions in which they received either an active alcohol-containing beverage (target breath alcohol concentration [BrAC] = 0.08 g/dL) or placebo. After absorption, pain threshold, suprathreshold pain intensity, and perceived pain relief were determined. For Study 1, pain threshold was assessed using a series of three slowly ramping thermal stimuli applied to the glabrous skin of the foot starting at 32 °C and increasing to a maximum of 50 °C at a rate of .5 °C/s. Participants indicated when sensation became painful. Suprathreshold pain intensity was assessed using three blocks of 7 thermal stimuli individually calibrated to produce a pain rating of approximately 50 out of 100. For Study 2, pain threshold was assessed using three slowly ramping pressure stimuli applied to the insertion of the masseter that increased at a rate of 0.5 lbf/s until participants indicated the sensation transitioned from pressure to pain. Suprathreshold pain intensity was assessed using 3 applications each of 4, 5, and 6 lbf stimuli to the masseter insertion, which are within the nociceptive range for most people. Participants self-reported pain intensity using 0–100 visual analog scales (VAS) anchored from “No pain at all” to “Most intense pain imaginable”. They also rated their perceived relief from pain as a result of consuming their beverage using a VAS anchored from “No relief at all” to “Most profound relief imaginable”. Pain thresholds, intensity ratings, and relief ratings were averaged for analysis purposes. Because pain threshold units differed between studies (°C vs. lbf), pain thresholds were Z-transformed for analysis purposes. 2 (Condition: Alcohol vs. Placebo) X 2 (Sex: Male vs. Female) X 2 (FH: Positive vs. Negative) mixed GLM including chronic pain status as a covariate was used to evaluate hypotheses.

Results: Participants averaged 26.5 (SD = 4.81) years of age and 17.2 years of education (SD = 2.14). Mean alcohol consumption over the past 6 months was .77 standard drinks per day. Alcohol Use Disorder Identification Test (AUDIT) scores averaged 4.53 (SD = 1.75), indicated minimal AUD symptomatology. Similarly, Beck Depression Inventory (BDI-II; $M = 3.05$, $SD = 3.89$) and State-Trait Anxiety Inventory (STAI; $M = 29.84$, $SD = 8.41$) were consistent with minimal affective disturbance. Mean BrAC during QST was 0.076 g/dL (SD = 0.016). For pain intensity, analyses revealed a significant condition X sex interaction ($F_{1,158} = 5.50$, $p = 0.02$, $2p = 0.034$) such that alcohol significantly decreased pain intensity for women ($p = 0.003$), but not men ($p = .71$). No other significant main or interactive effects were detected for pain intensity. For pain threshold, a significant condition effect was found such that alcohol significantly increased pain threshold vs. placebo ($F_{1,158} = 9.64$, $p = 0.002$, $2p = 0.058$). We also found a significant main effect of FH ($F_{1,158} = 11.58$, $p < 0.001$, $2p = 0.068$) qualified by a sex X FH interaction ($F_{1,158} = 12.58$, $p < 0.001$, $2p = 0.074$) such that FH- men had significantly higher pain thresholds than FH+ men ($p < 0.001$). No effect of FH on pain threshold was apparent for women ($p = 0.94$). Finally, ratings of perceived relief were significantly higher in the alcohol than

placebo condition ($F_{1,157} = 135.71$, $p < 0.001$, $2p = 0.46$). No other main or interactive effects achieved significance for perceived relief.

Conclusions: Results provided partial support for our hypotheses. Alcohol intake significantly increased pain threshold regardless of sex or FH. Alcohol also decreased suprathreshold pain intensity for women, but not men. Consistent with prior literature, these effects were small-to-moderate in magnitude. In contrast, alcohol intake produced very large increases in perceived pain relief compared to placebo. Contrary to hypotheses, FH did not significantly moderate alcohol effects on pain threshold, intensity, or relief. It is notable, however, that FH was associated with significantly greater pain sensitivity in men. Future studies should investigate whether FH of alcohol problems may act as a sex-specific risk factor for the development of chronic pain. Taken together, our findings suggest sex, but not FH, may moderate aspects of alcohol analgesia.

Keywords: Alcohol, Pain, Alcohol Subjective Response

Disclosure: Tryptamine Therapeutics, Consultant, Self, Oligomerix, Inc, Consultant, Self.

P289. TOP-DOWN REGULATION OF AMYGDALA AND STRIATUM WITH THETA-BAND STIMULATION OF FRONTOPARIETAL NETWORK IN OPIOID USE DISORDER

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Background: Theta-band frontoparietal synchronization is a promising way to boost the brain's control network and its regulation of reward regions important for recovery in addiction. In this pre-registered, triple-blind, sham-controlled trial (NCT03907644), we tested the first dual-site tACS-fMRI intervention targeting the right frontoparietal network for people with opioid use disorder (OUD).

Methods: Sixty male participants with OUD were randomized to receive 20 minutes of active or sham 6 Hz tACS (HD electrodes over F4 and P4). Structural, resting-state, task-based fMRI drug cue reactivity, and repeated cue-induced craving assessments were collected immediately before and after stimulation. Pre-registered outcome measures were analyzed using time X group interaction models to examine (1) modulation of drug cue-related brain activity, (2) changes in craving, (3) alterations in functional connectivity related to frontoparietal network, and (4) associations between electric field, neural responses, and craving behavior.

Results: Group by time generalized psychophysiological interaction analyses showed increased right frontoparietal network engagement ($\beta = 2.63$, $p = 0.0308$) following stimulation, and increased top-down inhibitory regulation of frontoparietal network on right ventral striatum ($\beta = 1.99$, $p = 0.037$) and left medial amygdala ($\beta = 1.97$, $p = 0.039$). Although subjective craving did not differ significantly between groups, group-specific analyses revealed a significant correlation between increased frontoparietal connectivity and reduced cue-induced craving in active ($r = -0.37$, $p = 0.05$), but not in the sham. Electric field strength in the right frontal node predicted frontoparietal network engagement in the active group ($r = 0.43$, $p = 0.02$).

Conclusions: Together, these findings provide the first causal evidence that theta-band frontoparietal tACS modulates cortical-subcortical circuits in OUD and supports network-targeted neuromodulation as a potential therapeutic intervention for addiction.

Keywords: Frontoparietal Network, Top down regulation, transcranial alternating current stimulation

Disclosure: Nothing to disclose.

P290. MEDICATION CLASS PRESCRIPTION RECEIPT BY INSOMNIA SYMPTOMS IN ADULTS RECEIVING BUPRENORPHINE FOR OUD

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Background: Sleep disturbance is common among individuals with opioid use disorder (OUD) receiving buprenorphine treatment and are associated with poor OUD treatment outcomes. Recent administrative claims studies indicate that over half of adults on buprenorphine report clinically significant insomnia symptoms and are frequently prescribed sedative/hypnotic medications, which can bring additional risks without clear benefit. Yet it is unclear whether use of different medication classes are differentially prescribed to patients by their reported specific insomnia symptoms in clinical practice. This study compares medication prescription frequencies between participants who endorsed versus did not endorse key insomnia symptoms (difficulty falling asleep, difficulty staying asleep, and waking up too early) among a sample of patients receiving outpatient OUD treatment with buprenorphine.

Methods: This study was a cross-sectional analysis of a patient-administered survey and medical record abstraction from a sample of adults with OUD receiving buprenorphine at an academic addiction clinic (N = 136). The Insomnia Severity Index (ISI) was used to classify endorsement of three symptoms typically used to categorize insomnia types – difficulty falling asleep, difficulty staying asleep, and waking up too early – each dichotomized as endorsed (moderate/severe) versus not endorsed (none/mild). Medical record abstracted prescriptions were grouped into four classes: benzodiazepines; Z-drugs (non-benzodiazepine hypnotics); melatonergic agents (melatonin, ramelteon); and other medications which are known to commonly be prescribed for sleep in real-world clinical practice (SSRIs, trazodone, mirtazapine, gabapentin, quetiapine, hydroxyzine). For each symptom × medication class, 2 × 2 contingency tables (symptom endorsed vs not; ≥1 medication in class present vs absent) were constructed. Frequencies (%) were summarized, and differences in proportions were tested using Pearson's chi-square or Fisher's exact tests (two-sided), as appropriate. Odds ratios (95% CI) were calculated using the Baptista–Pike method.

Results: Our sample included 136 adults (mean age 40.0 ± 10.0 years; 59.6% women). Mean duration of buprenorphine treatment was 1.86 years (1.61); 88.2% were receiving sublingual formulation at a median sublingual dose of 20 mg/day (IQR 16–24) and Sublocade dosing was 300 mg in n = 13 and 100 mg in n = 3 (Table 1). Frequencies by symptom endorsement were as follows: the 'Other' class (SSRIs, trazodone, mirtazapine, gabapentin, quetiapine, hydroxyzine) was most frequently prescribed across symptom endorsement; for difficulty falling asleep, 'Other' medications were prescribed to 78.6% of endorsers vs 63.6% of non-endorsers; for difficulty staying asleep, benzodiazepines were prescribed to 4.9% vs 16.7%; for waking too early, Z-drugs were infrequently prescribed in both endorsement groups. Two-sided Pearson's chi-square or Fisher's exact tests were used, where applicable. For difficulty staying asleep vs benzodiazepine use: $\chi^2(1) = 5.23$, $p = 0.022$; OR = 0.26 (95% CI 0.07–0.88). Full percentages, odds ratios (95% CIs), and two-sided p-values for each symptom–medication pairing appear in Table 2.

Conclusions: Medication prescription patterns differed by ISI symptom endorsement. The 'Other' class (SSRIs, trazodone, mirtazapine, gabapentin, quetiapine, hydroxyzine) was most frequently reported overall, whereas Z-drugs and melatonergic agents were uncommon. Benzodiazepine use was less frequent

among those endorsing difficulty staying asleep than among non-endorsers. These data provide symptom-specific frequency estimates and motivate future work to clarify indications, prescribing decisions, and outcomes in buprenorphine-treated OUD populations with co-morbid insomnia.

Keywords: opioid use disorder, insomnia, medications, Buprenorphine

Disclosure: Nothing to disclose.

P291. CO-USE OF ALCOHOL AND OPIOID USE FOLLOWING PHYSICAL PAIN IN YOUNG ADULTS IN THE NATURAL ENVIRONMENT

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Background: Alcohol and opioids both have analgesic effects that occur, in part, through activation of the endogenous opioid system. People who use both substances together may experience dysregulation of pain neurocircuitry and neurochemistry. However, minimal work has examined alcohol-opioid co-use. This presentation will examine whether physical pain plays a role in the co-use of these two substances, a dangerous combination that increases overdose risk over either substance alone.

Methods: Participants were young adults aged 18–26 (N = 30; men = 15, women = 15) who reported alcohol-opioid co-use at least weekly. Participants were not recruited based on their experience of pain. They completed 21 days of ecological momentary assessment (EMA), answering via smartphone 4 self-report surveys per day at random times, self-initiating reports when they began using alcohol and/or opioids, and completing follow-ups over the two hours following their initial use (total obs = 2843). Participants reported their current pain and pain catastrophizing at all reports. Multilevel logistic and linear models, which account for the nesting of EMA surveys within participants, were used to examine associations of pain with subsequent substance use and vice versa. Multilevel multinomial logistic regression was used to examine differences between alcohol use, opioid use, and alcohol-opioid co-use. All analyses were conducted within-person, leveraging the fact that we had approximately 95 observations per person (2843 total observations).

Results: Participants initiated 61 alcohol-only, 163 opioid-only, and 96 alcohol-opioid co-use episodes. They reported pain on 18% of non-substance use occasions and during 15% of use episodes. Relative to alcohol-only episodes, pain was elevated at the start of opioid (OR = 1.41, CI = [1.09, 1.82], $p = 0.009$) and alcohol-opioid episodes (OR = 1.33, CI = [1.03, 1.71], $p = 0.028$). In time-lagged analyses, previous-occasion pain interacted with pain catastrophizing to specifically predict alcohol-opioid co-use, such that, at moments following elevated pain, catastrophizing was associated with greater odds of co-use (OR = 1.45, CI = [1.04, 2.00], $p = 0.027$). Following use initiation, pain decreased over follow-ups ($b = -0.28$, CI = [−0.43, −0.13], $p < 0.001$).

Conclusions: Despite not being recruited based on pain experience, participants frequently reported pain. Findings support negative reinforcement addiction models, with participants using opioids when experiencing pain and reporting pain relief following use. This may lead to increased use over time as tolerance develops. Co-use of alcohol and opioids was particularly likely when participants were concerned by their pain. Pain and pain catastrophizing may be key intervention targets for preventing the dangerous combination of alcohol and opioids.

Keywords: Pain, opioid, Alcohol

Disclosure: Nothing to disclose.