POSTER ABSTRACT 1

William Reay
School of Biomedical Sciences and Pharmacy, The University of Newcastle, Australia

Clinically Actionable Pathways Identified in Individuals With Schizophrenia by Pharmacological Enrichment of Polygenic Risk

Background: Psychiatric disorders are complex traits that remain difficult to effectively treat. While the heritable burden of common variation can be expressed in individuals as a polygenic risk score (PRS), it is not directly informative for pharmacotherapy due to its composition of heterogenous risk factors genome-wide. We therefore aimed to develop a framework to quantify an individual's polygenic risk in systems responsive to existing drugs.
Methods: We developed the pharmagenic enrichment score (PES) to quantify common variation in systems which interact with approved drugs. The PES framework identifies clinically actionable pathways enriched with common variation using GWAS summary statistics. Polygenic risk specifically for these systems (PES) can then be profiled in individual patients for precision drug repositioning. This approach was applied to schizophrenia using GWAS summary statistics and a genotyped cohort of schizophrenia cases.
Results: Using schizophrenia GWAS data we identified eight candidate gene-sets with known drug targets which displayed common variant enrichment in schizophrenia. A large proportion of cases in our study cohort had elevated PES in one or more of the eight clinically actionable gene-sets. Notable compounds targeting these pathways included vitamins, insulin modulating agents, and protein kinase inhibitors with putative neuroprotective properties. Interestingly, individual PES profiles were not significantly associated with genome-wide schizophrenia PRS, with elevated PES observed in patients with otherwise low schizophrenia polygenic risk.
Conclusion: Our data suggest that the PES framework could integrate an individual's common variant risk to inform personalised interventions, including drug repositioning, for complex disorders such as schizophrenia.

POSTER ABSTRACT 2

Susan Shen
University of California San Francisco

A Candidate Causal Variant Underlying Both Enhanced Cognitive Performance and Increased Risk of Bipolar Disorder

Background: Bipolar disorder is a highly heritable mental illness, but the underlying genetic and molecular mechanisms are largely unknown. Recent GWAS's have identified an intergenic region associated with both cognitive performance and bipolar disorder. This region contains dozens of putative fetal brain-specific enhancers and is located ~0.7 Mb upstream of the neuronal transcription factor POU3F2.
Methods and Results: We identified a candidate causal variant, rs77910749, that falls within a highly conserved putative enhancer, LC1. This human-specific variant is a single-base deletion in a PAX6 binding site and is predicted to be functional. We hypothesized that rs77910749 alters LC1 activity and hence POU3F2 expression during neurodevelopment. Indeed, transgenic reporter mice demonstrated LC1 activity in the developing cerebral cortex and amygdala. Furthermore, ex vivo reporter assays in embryonic mouse brain and human iPSC-derived cerebral organoids revealed increased enhancer activity conferred by the variant. To probe the in vivo function of LC1, we deleted the orthologous mouse region, which resulted in amygdala-specific changes in Pou3f2 expression. Lastly, 'humanized' rs77910749 knock-in mice displayed behavioral defects in sensory gating, an amygdala-dependent endophenotype seen in patients with bipolar disorder.
Conclusion: Our study suggests a molecular mechanism underlying the long-speculated link between higher cognition and neuropsychiatric disease.

POSTER ABSTRACT 3

Gil Hoftman
Semel Institute for Neuroscience & Human Behavior at UCLA

Association of Primate-Specific Supragranular Enriched Gene Expression With Cortical Thickness Patterns in 22q11.2 Deletion Syndrome

Background: The 22q11.2 deletion syndrome (22q11DS) typically results from a 1.5-3 megabase deletion on chromosome 22, has a broad multi-organ system phenotype, and spans up to 46 protein coding genes. However, how specific genes outside the deletion area may interact with 22q11DS genes that are associated with spatial neuroanatomical hierarchy requires further study. Here, we examined a set of genes enriched in human supragranular layers 2 and 3 (hSEG) and asked whether expression levels are associated with a cross-cortical region deviance pattern in cortical thickness in 22q11DS.
Methods: Neuroanatomic alterations using structural MRI measures were characterized in a sample of 232 subjects with 22q11DS and demographically matched comparison subjects (n=290). Transcriptional data from the Allen Human Brain Atlas was used to systematically analyze the spatial pattern of each hSEG (n=19) and the pattern of regional cortical thickness deviance in 22q11DS.
Results: Eight of the 19 hSEG genes had spatial expression patterns that were strongly associated with cortical thickness deviance in 22q11DS. Interestingly, four of these genes (NEFH, SCN4B, SYT2 and VAMP1) have a gradient of increasing levels across the rostrocaudal axis. Furthermore, these four genes were highly correlated with P2RX6 and AIFM3, both within the 22q11 region that were strongly associated with cortical thickness deviance.
Conclusion: Mapping gene expression within and outside the 22q11.2 deletion area with non-invasive neuroanatomical measurements, particularly those genes enriched in primate supragranular layers, may produce data important for generating hypotheses about circuit- and molecular-specific changes in 22q11DS.

POSTER ABSTRACT 4

Evan Geller
Yale University School of Medicine

High-Throughput Disruption of Enhancers Active During Human Corticogensis

Genetic variation within developmental regulatory elements are thought to contribute neurodevelopmental disorders. We hypothesized that identification of regulatory elements which alter human neural stem cell (hNSC) proliferation will provide insight into the genetics of neurodevelopment. Here we implement a high-throughput method to interrogate more than 25,000 individual binding sites across thousands of enhancers active in the developing human cortex and more than 9,500 expressed genes. Specifically, we utilized active sgRNA-Cas9 to 'disrupt' conserved elements within enhancers and to knockout protein-coding genes.

We identified 1,192 unique deletions within enhancers and 2,264 gene knockouts that alter hNSC proliferation. By 12 cell divisions, a distinct set of conserved regions within enhancers decrease proliferation (3.9%) or increase proliferation (0.8%). These binding sites are enriched for specific transcription-factor motifs, including E2F4, SP1, and KLF5, suggesting functions in cell cycle regulation, local chromatin remodeling, and response to extrinsic developmental cues. Intriguingly, we also identify enhancer disruptions directly interacting with known neurodevelopmental risk genes.

As expected, we identified many gene knockouts that decrease proliferation (22.9%) and a smaller set of gene knockouts that increase proliferation (<0.1%). Proliferation decreasing genes are notably enriched for ASD and developmental disorder risk genes, providing insight into how loss-of-function mutations in these genes may contribute to disease risk. Our results provide quantitative maps of genes and regulatory elements affecting hNSC proliferation and reveal novel insights into enhancer function during development. We expect these findings to aid the interpretation of non-coding genetic variation linked to developmental disorders.

POSTER ABSTRACT 5

Huei-Ying Chen
Lieber Institute for Brain Development

Loss of Inhibitory Neurons and Perturbed Inhibition in a Syndromic ASD Mouse Model

Pitt-Hopkins Syndrome (PTHS) is a neurodevelopmental disorder with autistic features caused by autosomal dominant mutations in the transcription factor 4 (TCF4) gene. The pathophysiology underlying PTHS remains unresolved. In situ hybridization studies indicate Tcf4 is enriched in the proliferative zone of the embryonic ventral forebrain during neurogenesis of GABAergic interneuron, and Tcf4 is also known to dimerize with Ascl1, a gene critical for interneuron development. Using a PTHS mouse model, we found that prefrontal pyramidal neurons show a reduction in the frequency of spontaneous inhibitory currents, suggesting mutations in Tcf4 lead to a deficit in cortical inhibition. To test the hypothesis that Tcf4 regulates interneuron development and function, we first examined interneuron numbers in the PTHS mouse. We immunostained for parvalbumin (PV) and somatostatin (SST), two GABAergic cell markers, and found a significant decrease in the PV population in the medial prefrontal cortex, primary motor cortex, striatum, basal lateral amygdala, and the hippocampus. This deficit appears to be specific to PV-positive cells, as preliminary results suggest the SST population is unaffected. Loss of GABAergic neurons was further validated by crossing tdTomato reporter line with cortistatin-Cre transgenic, in which mixed classes of GABAergic interneurons are labeled. Despite the fact that the PV population is reduced, the intrinsic membrane properties of the remaining PV-positive interneurons appear to be electrophysiologically normal. Together, these results suggest that inhibitory tone in the PTHS mouse is perturbed, and these changes may in part underlie the cognitive deficits and autistic features present within this patient population.

POSTER ABSTRACT 6

Gwynne Davis
Institute for Neurodegenerative Diseases UCSF

The Effect of Ketamine on Fronto-Striatal Circuit Mechanisms Underlying Compulsive Grooming Behavior

Obsessive-Compulsive Disorder (OCD), characterized by intrusive thoughts (obsessions) and repetitive behaviors (compulsions), is associated with dysfunction in fronto-striatal circuits. Animal studies find fronto-striatal activity modulation is a critical driver of OCD-like repetitive behaviors. Additionally, genetic disruptions that alter excitatory fronto-striatal synapses result in OCD-like behavior. An intravenous infusion of ketamine, an NMDA receptor antagonist, can rapidly reduce obsessive thoughts in adults with OCD. To probe mechanisms underlying ketamine's therapeutic effect on OCD-like behaviors, we used the SAPAP3 knockout (KO) mouse model of compulsive grooming. We injected SAPAP3 KO mice and wild-type (WT) littermates with either saline or ketamine (30 mg/kg) and measured their grooming behavior for 7 days post-injection. Ketamine significantly attenuated grooming behavior in KO mice compared to saline controls. Histological analysis revealed that KO mice had reduced cFos+ cells in the prelimbic cortex (PL) under saline conditions, and that ketamine increased cFos+ cells in the dorsal medial striatum (DMS). We performed in vivo fiber photometry recordings from PL-DMS projection neurons 24 hours post-injection. Compared to saline, ketamine increased the amplitude of calcium transients in KO mice during grooming epochs. Optogenetically increasing activity in PL-DMS projections was sufficient to rescue the compulsive grooming phenotype in KO mice. Conversely, optogenetic inhibition of PL-DMS projections increased grooming in WT mice, demonstrating bidirectional control of grooming behavior via this fronto-striatal circuit. These studies demonstrate that ketamine increases activity in a fronto-striatal circuit that causally controls compulsive grooming behavior, suggesting this circuit may be important for ketamine's therapeutic effects in OCD.

POSTER ABSTRACT 7

Himanshu Mishra
Department of Psychiatry and Center for Circadian Biology, University of California San Diego; Psychiatry Service, Veterans Affairs

Human Induced Pluripotent Stem Cell Derived Neurons Model Circadian Rhythm Abnormalities in Bipolar Disorder

Background: Bipolar disorder (BD) is a neuropsychiatric condition characterized by recurrent mood episodes, suicidal thoughts and altered daily rhythms in activity, energy, cognition and appetite. These symptoms suggest that circadian rhythms may be disrupted but the underlying molecular mechanisms are not well understood. Recent advancements in reprogramming technologies have enabled the use of induced pluripotent stem cells (iPSCs) as powerful tools for investigating the relationships between genotype and phenotype in human neuropsychiatric disease-relevant cells.
Methods: We used a set of ten iPSC lines from six BD patients and four age-matched controls. iPSCs were differentiated into early neuronal progenitors (NPCs) and forebrain VGlut1+ neurons. We then examined temporal differences in clock gene expression and stage-specific circadian rhythm profiles in control and BD neural cells.
Results: Time course qPCR revealed higher PER2 expression in NPCs and neurons from BD patients compared to controls. Bioluminescent reporter Per2-luc activity exhibited robust rhythms in controls and weaker rhythms in BD-NPCs. Circadian period was shorter in cells from BD patients, especially in lithium-responder group. Single cell imaging analysis corroborated weaker rhythms in BD. Differentiated neurons in BD showed significantly decreased amplitude. Interestingly, rhythms were weaker and dampened faster in BD neurons compared to controls.
Conclusions: Our data indicate that neural cells show aberrant rhythms in BD. Our patient derived neuronal model of circadian rhythms is an ideal platform for future studies to define new targets for pharmacological modulation of cellular rhythms in NPCs and neurons from bipolar disorder.

POSTER ABSTRACT 8

Perry Spratt
University of California San Francisco

Crispr Activation Rescues Physiological Deficits Associated With SCN2A Haploinsufficiency

De novo mutations in the gene SCN2A are strongly associated with autism spectrum disorder (ASD) and intellectual disability. The majority of ASD-associated SCN2A mutations are protein truncating variants, resulting in individuals having only one functional copy of SCN2A (haploinsufficiency). SCN2A encodes NaV1.2, a voltage-gated sodium channel expressed throughout the brain. Using a mouse model heterozygous for Scn2a, we found that NaV1.2 loss resulted in developmentally distinct deficits in prefrontal cortex excitatory neurons. Action potential initiation was impaired in early in development, while deficits in dendritic excitability persisted throughout life. These deficits were associated with impaired excitatory synapses, even when Scn2a was disrupted late in development. These findings suggest that NaV1.2 function is critical throughout life, raising the possibility that restoring normal NaV1.2 function, even later in development, may result in a therapeutic benefit for individuals with ASD-associated SCN2A mutations. To explore this possibility, we have developed CRISPR activation (CRISPRa) tools to increase the expression of the remaining functional SCN2A allele to normal physiological levels. CRISPRa targets a transcriptional activator to regulatory elements of individual genes, upregulating their transcription. Using this approach, we optimized a recombinant adeno-associated virus-based system that upregulates Scn2a by targeting its promoter. Injecting this viral system into the prefrontal cortex increased Scn2a expression and restored features of intrinsic excitability and synaptic transmission. This suggest that restoring Scn2a expression can rescue cellular deficits associated with Scn2a haploinsufficiency and that CRISPRa could be utilized as a potential therapeutic for other haploinsufficient genes in ASD and additional neurodevelopmental conditions.

POSTER ABSTRACT 10

Jae Hoon Sul
Department of Psychiatry and Biobehavioral Sciences, University of California Los Angeles

Contribution of Common and Rare Variants to Bipolar Disorder Susceptibility in Extended Pedigrees From Population Isolates

Current evidence from case/control studies indicates that genetic risk for psychiatric disorders derives primarily from numerous common variants, each with a small phenotypic impact. The literature describing apparent segregation of bipolar disorder (BP) in numerous multigenerational pedigrees suggests that, in such families, large-effect inherited variants might play a greater role. To identify roles of rare and common variants on BP, we conducted genetic analyses in 26 Colombia (CO) and Costa Rica (CR) pedigrees ascertained for bipolar disorder 1 (BP1), the most severe and heritable form of BP. In these pedigrees, we performed microarray SNP genotyping of 838 individuals and high-coverage whole-genome sequencing of 449 individuals. We compared polygenic risk scores (PRS) using the latest BP1 genome-wide association study (GWAS) summary statistics between BP1 individuals and related controls. We also evaluated whether BP1 individuals had a higher burden of rare deleterious single nucleotide variants (SNVs) and rare copy number variants (CNVs) in a set of genes related to BP1. To identify rare variants that segregated with BP1 in the pedigrees, we developed a new statistical approach that calculates segregation p-values for rare variants. We found that compared to unaffected relatives, BP1 individuals had higher PRS estimated from BP1 GWAS statistics and displayed higher burdens of rare deleterious SNVs and rare CNVs in genes related to BP1. We observed no significant segregation pattern for rare variants. These results suggest that small to moderate effect rare and common variants contribute to BP1 risk in extended pedigrees.

POSTER ABSTRACT 11

Min Woo Sun
Stanford University

Game Theoretic Centrality Supports Link Between Autism Spectrum Disorder and the HLAComplex

Background: Complex genetic diseases with etiological heterogeneity like Autism Spectrum Disorder (ASD) often pose a challenge for traditional genome-wide association study approaches in defining a clear genotype to phenotype model. We propose a novel method based on coalitional game theory that can capture the combinatorial interaction of mutations that manifests into a disease.
Methods: Shapley value is a popular solution for coalitional games that measures the average marginal contribution of a player—in our case genes—across all possible coalitions. Game centrality extends the notion of Shapley value to the evaluation of a gene's contribution to the overall connectivity of its corresponding node in a biological network. Each of the nodes can also be assigned a weight based on a priori importance of the genes.
Results: We applied game centrality to genomes from 756 multiplex autism families. Likely, gene disrupting mutations in coding regions were encoded into case (ASD) and control (unaffected) binary matrices and the corresponding genes were used to generate a protein-protein interaction graph using STRING. The top five percent of genes with the highest game centrality were enriched for pathways of the immune system. In particular three of the selected genes are part of the human leukocyte antigen complex (HLA), which has been previously associated with ASD.
Conclusion: Despite the evidence for strong genetic etiology, the exact genotype responsible for the varying ASD phenotypes remains unclear. Game centrality can facilitate the identification of influential, disease-associated genes within biological networks, thereby decoding the polygenic underpinnings of diseases like autism.

POSTER ABSTRACT 12

Karol Cichewicz
Center for Neuroscience, University of California Davis

Maternal Immune Activation Perturbs Transcriptional Co-Expression Networks During Corticogenesis

Maternal immune activation (MIA) has emerged as an environmental risk factor for neurodevelopmental disorders (NDDs). Animal models of MIA provide an opportunity to identify molecular pathways that initiate disease processes and lead to neuropathology and behavioral deficits in offspring. Here, we applied transcriptional profiling and neuroanatomical characterization across embryonic cortical development following MIA via viral mimic polyinosinic:polycytidylic acid. MIA induced strong transcriptomic signatures, including induction of genes associated with hypoxia, immune signaling, and angiogenesis. The acute response was followed by precocious changes in proliferation, neuronal and glial differentiation, and cortical lamination that emerged at E14.5 and peaked at E17.5. MIA-induced transcriptional changes were largely suppressed by maternal IL-6 inhibition. MIA transcriptomic signatures overlap significantly with perturbations identified in NDDs and provide novel insights into molecular and developmental processes linking MIA and neuropathology, potentially revealing new targets for development of novel approaches for earlier diagnosis and treatment of these disorders.

POSTER ABSTRACT 13

Cesar Canales
University of California Davis

Initial Insights Into the Role of CHD8 in the Cerebellum

Mutation in the chromatin-remodeling factor CHD8 (Chromodomain-Helicase DNA Binding Protein 8) have emerged as a key genetic risk factor for neurodevelopmental disorders (NDDs) and autism spectrum disorder (ASD). Most CHD8 mutations lead to a loss of functions and carrier patients display, among other physiological malfunctions, intellectual disability, ASD-like behavior and macrocephaly.
Chd8 haploinsufficiency in mice (Chd8+/del5bp) presents similar ASD-like patterns, altered proliferative cortical dynamics, and MRI studies have determined that Chd8+/del5bp mice have slightly smaller than average size cerebellums amongst other anatomical brain alteration.
The cerebellum is the region of the brain largely responsible for motor control and coordination and recent studies have highlighted the potential of this region as a model to study cell fate determination and postmitotic maturation of a single predominant neuronal cell type over the full-time course of differentiation.
Here, in order to understand the causal roots of the MRI findings in Chd8+/del5bp, and link our analysis to cerebellar function and alterations in proliferative dynamics previously described in the developing cortex, we have performed transcriptional profiling in cerebellum and studied cerebellar neuroanatomy through immunohistochemistry (IHC), looking at distribution of Purkinje cell neurons and proliferative dynamics. We have also performed behavioral motor-coordination and developmental milestone phenotyping. Altogether, this data provides new insights into the function of CHD8 in the cerebellum, an ASD-relevant brain region that is often overlooked across NDD animal models.

POSTER ABSTRACT 14

Kaitlin Scarborough
Des Moines University

Choreiform Movements in Methamphetamine-Induced Movement Disorder: A Rare but Recognized Presentation

Choreiform movement disorder associated with methamphetamine abuse is a rarely documented problem. At least 13 cases have been reported, and this poster presentation adds to that body of literature. Considered by some authors to be the most popular psychostimulant in the world, methamphetamine is a major drug of abuse in the United States. Methamphetamine displaces monoamines, inducing increased vesicle release of norepinephrine at lower doses, and dopamine at moderate doses at the pre-synaptic terminal. It also stimulates release of serotonin at higher doses, and acts as a direct agonist of 5-HT receptors. The pathophysiology of abnormal movements in substance-induced movement disorder, Huntington disease, and Parkinson disease share a theme of perturbations within the basal ganglia. The presence of choreiform movements in some methamphetamine abusers is not surprising, given the evidence that dopamine terminals are damaged in the caudate (more prevalent) and putamen (less prevalent) in chronic methamphetamine users. The interactions amongst norepinephrine, dopamine, GABA, and acetylcholine within the basal ganglia, while well-characterized, are still incompletely understood. The basal ganglia are implicated in the permission of desired movements and the inhibition of undesired ones. Hyperkinetic movements occur secondary to disinhibition of the pallidum, yet even this is an oversimplification as the pallidum is involved with both the direct (GABAergic) and indirect (GABAergic and glutamatergic) pathways. Our poster describes the presentation and treatment of a patient with choreiform movements associated with methamphetamine abuse, along with a discussion of previously documented treatment options and a proposed approach to treatment algorithm.

POSTER ABSTRACT 15

Diana Waters
Duke University, Department of Neurobiology

Developing a Behavioral Task to Assay Positive and Valence Systems in a Bipolar Disorder Mouse Model

Balance between the positive and negative valence systems is necessary to appropriately compare risk versus reward when completing a motivated behavior. The manic behaviors of bipolar disorder (BPD) can be framed as an improper balance of the brain's positive and negative valence systems. In mania, there is an increase in positive valence states with enhancements in motivation and reward-based decision-making as well as a decrease in negative valence states with impairments of fear and anxiety responses. To better understand the balance of positive and negative valence systems, we developed a behavioral task modeled after the classic elevated plus maze and sucrose preference tasks. The Approach-Avoidance Task, performed in 2 phases, directly quantifies the impact of anxiogenic stimuli on reward-motivated behavior. In the first phase of the task, The Serial Reward Task, animals are free to consume a sucrose solution from 4 delivery sites. In the second phase, The Approach-Avoidance phase, anxiogenic stimuli are introduced en route to the reward sites. By utilizing CLOCKΔ19 mice, an established mouse model of mania, we found that CLOCKΔ19 mice have increased sucrose reward drive and displayed decreased sensitivity to anxiogenic stimuli compared to wildtype littermates. This validated behavioral task, along with chronic in vivo electrophysiology, has the potential to distinguish the differences in circuit dynamics that underly these behaviors in wildtype animals and models of BPD.

POSTER ABSTRACT 16

Peter Washington
Stanford University

Data-Driven Identification of Predictive Social Responsiveness Biomarkers for Autism

Background: Autism Spectrum Disorder (ASD) is a complex neuropsychiatric condition with a highly heterogeneous phenotype. The Social Responsiveness Scale (SRS) is a 65-item questionnaire that is often used to assess ASD severity. We perform item-level question selection on answers to the SRS to determine whether autism can be classified with a small subset of questions.
Methods: We aggregated 8 databases which collectively contain the SRS answers corresponding to 16,527 ASD cases and controls. We perform filter, wrapper, and embedded feature selection analyses to identify top-ranking questions. We then compress the 65-question SRS into low-dimension representations using PCA, t-SNE, and a denoising autoencoder. Using a multi-layer perceptron classifier and 10-fold cross validation, we measure the performance of a reduced feature set consisting of only top-ranking questions for different set sizes. We also evaluate feature sets consisting of varying sizes of lower dimension representations of the data.
Results: Trouble relating to peers is the top-rated feature for predicting the SRS-derived diagnosis, while trouble keeping up with conversation flow is the top-rated feature for predicting the dataset-provided diagnosis. Classification using only the top-rated question resulted in an AUC of 91%-93% for SRS-derived diagnoses and an AUC of 80%-84% for dataset-specific diagnoses. Single dimensional representations of the data across all three methods resulted in an AUC over 99% for SRS-derived diagnosis and 87% for dataset-specific diagnosis.
Conclusion: Autism can be classified with high accuracy using a small subset of behavioral features.

POSTER ABSTRACT 17

Rinaldo Catta-Preta
University of California Davis

On the Mechanisms of DLX Transcriptional Regulation of Critical Genes in Basal Ganglia Development and Interneuron Specification

Interneurons modulate cortical signaling, and perturbance to proliferation, migration and specification of cortical interneurons (CIN) are implicated in neurodevelopmental disorders (NDDs). Transcription factors control the development of cell types such as CIN via organizing gene regulatory networks. Dlx genes have been shown to be master TFs that control gene expression patterns required for early basal ganglia development and CIN specification, yet genomic function of DLX TFs has yet to be defined.
We conducted RNA-seq and ChIP-seq experiments on embryonic basal ganglia in wild-type and a Dlx1/2-/- mutant mice. We computationally integrated genomic, epigenomic and neuroanatomical results to characterize the regulatory wiring in early basal ganglia development orchestrated by DLX2, DLX1 and DLX5.
DLX TFs interact with the genome broadly, with the binding patterns of DLX2, DLX1, and DLX5 largely overlapping and expanding from E11.5 to E16.5 in the embryonic ganglionic eminences. A small subset of DLX-bound loci that were sensitive to DLX ablation represent a coherent regulatory network of genes that are either activated and drive GABAergic specification or repressed to drive maturation from early developmental states and repress alternative fates.
Our results reveal DLX regulatory function and explain the paradox of widespread TF binding contrasted with significant epigenomic and transcriptomic changes only in a limited subset of target genomic interactions. The regulatory properties identified for DLX TFs suggest general mechanisms by which TFs orchestrate dynamic expression programs underlying neurodevelopment.

POSTER ABSTRACT 18

Aaron Jenkins
University of Pittsburgh, Department of Psychiatry

Increased Markers of Microglia and Complement-Mediated Pathways in Schizophrenia

Background: Schizophrenia (SZ) is a severe psychiatric disorder associated with cognitive disturbances linked to dysfunction within the prefrontal cortex (PFC). Genetic studies have implicated microglia-related genes as risk factors for the disorder; however, the specific contribution of these cells to the underlying pathophysiology is unknown. Emerging evidence demonstrates that microglia are involved in the phagocytosis of dendritic spines and may exert their effects through complement-mediated processes. Given well-known evidence of spine deficits within the disorder, these findings suggest that microglia may contribute to these losses.
Methods: Quantitative PCR was used to assess transcript levels of markers that are enriched within microglia as well as components of the complement pathway that mediate phagocytosis in the PFC of 62 matched pairs of SZ and unaffected comparison subjects.
Results: Analyses revealed significantly increased transcript levels of multiple microglial markers involved in phagocytosis, including CD68 and the TAM receptor tyrosine kinases Axl and MerTK, in SZ subjects relative to comparison subjects. Additionally, several components of the complement pathway, including C1q and C4 were also elevated.
Conclusions: These results suggest that SZ subjects have higher levels of critical molecular machinery that supports microglial phagocytosis in the PFC and consequently may play a role in spine deficits within the disorder. Future studies that investigate the relationship between microglia-mediated phagocytosis, alterations within the complement pathway, and spine loss may help elucidate specific mechanisms underlying cognitive dysfunction within SZ and guide the identification of novel pharmacologic targets.

POSTER ABSTRACT 19

Xiao-Hong Lu
LSUHSC

Dosage Sensitivity Intolerance of VIPR2 Microduplication is Disease Causative to Manifest Schizophrenia-Like Phenotypes in a Novel BAC Transgenic Mouse Model

Background: Recent genome-wide association studies (GWAS) have identified copy number variations (CNVs) at chromosomal locus 7q36.3 that significantly contribute to the risk of schizophrenia, with all of the microduplications occurring within a single gene: Vasoactive intestinal peptide receptor 2 (VIPR2).
Methods: To confirm disease causality and translate such a genetic vulnerability into mechanistic and pathophysiological insights, we developed a series of conditional VIPR2 Bacterial Artificial Chromosome (BAC) transgenic mouse models of VIPR2 CNV. The conditional design of the BAC allows switching-off the transgene expression in desired spatiotemporal patterns.
Results: VIPR2 CNV mouse model recapitulates gene expression and signaling deficits seen in human CNV carriers. VIPR2 microduplication in mice elicits dorsal striatal dopamine dysfunction, cognitive, sensorimotor gating and social behavioral deficits preceded by an increase of striatal cAMP/PKA signaling and the disrupted early postnatal striatal development. Genetic removal of VIPR2 transgene expression via crossing with Drd1a-Cre BAC transgenic mice rescued the dopamine D2 receptor abnormality and multiple behavioral deficits, implicating a pathogenic role of VIPR2 overexpression in dopaminoceptive neurons.
Conclusion: Thus, our results provide further evidence to support the GWAS studies that the dosage sensitivity intolerance of VIPR2 is disease causative. The conditional BAC transgenesis offers a novel strategy to model CNVs with a gain-of -copies and facilitate the genetic dissection of when/where/how the genetic vulnerabilities affect development, structure, and function of neural circuits. Our findings have important implications for therapeutic development, and the etiology-relevant mouse model provides a useful preclinical platform for drug discovery.

POSTER ABSTRACT 20

OPEN BOARD

POSTER ABSTRACT 21

Bashkim Kadriu
Division Intramural Research Program, National Institute of Mental Health

The Impact of Ketamine and AV-101 on the Kynurenine Pathway in Subjects With Treatment-Resistant Unipolar or Bipolar Depression

Background: Glutamatergic dysfunction and immune system dysregulation are both thought to be involved in the pathophysiology of depression. One potential convergence point for these systems is the kynurenine (KYN) pathway. Pathological activation of the KYN pathway via rate-limiting enzyme indoleamine-2,3-dioxygenase (IDO) affects two critical downstream byproducts—kynurenic acid (KynA) and quinolinic acid (QA)—that trigger microglial activation, thereby altering glutamate release/reuptake. Here we assessed the effect of single-dose ketamine and/or two weeks of oral AV-101 impact on key components of the KYN pathway, in subjects with treatment-resistant bipolar depression (BD) or major depressive disorder (MDD).
Method: Data were drawn from two separate double-blind, randomized trials assessing the efficacy of single-dose ketamine (0.5 mg/kg) in subjects BD (n=39) or two weeks of oral AV-101 (1080 or 1440mg/day) in MDD subjects (n=19). ELISA tests were used to characterize key components of the KYN pathway pre-(baseline) and post-ketamine (230min, Day1, and Day3 post-ketamine) or AV-101 (assessed at baseline through Day13 post-AV-101).
Results: Single-dose ketamine altered key components of the KYN pathway on BP subjects. This included significantly decreasing IDO levels and increasing levels of KYN and KynA. In contrast, two weeks of oral AV-101 had no effects on KYN pathway measures or other central biological indices, including imaging measures.
Conclusion: Taken together, the results demonstrate that, in addition to having rapid and sustained antidepressant effects in BD subjects, ketamine affects key components of the KYN pathway. Unlike ketamine, AV-101 had no significant treatment efficacy MDD and no significant effects on the KYN pathway.

POSTER ABSTRACT 23

Sara Linker
Salk Institute for Biological Studies

Identifying a Predictive Signature of Neuronal Reactivity in Single Hippocampal Nuclei

Activity-induced remodeling of neuronal circuits is critical for memory formation. This process relies in part on transcription, but neither the rate of activity nor baseline transcription is equal across neuronal cell types. In this study, we isolated mouse hippocampal populations with different activity levels and used single nucleus RNA-seq to compare their transcriptional responses to activation. We found that 1 hr after novel environment exposure sparsely active dentate granule (DG) neurons had a much stronger transcriptional response compared to more highly active CA1 pyramidal cells and vasoactive intestinal polypeptide (VIP) interneurons. Activity continued to impact transcription in DG neurons up to 5 hr, with increased heterogeneity. By re-exposing the mice to the same environment, we identified a unique transcriptional signature that selects DG neurons for reactivation upon re-exposure to the same environment. These results link transcriptional heterogeneity to functional heterogeneity and identify a transcriptional correlate of memory encoding in individual DG neurons.

POSTER ABSTRACT 24

Maya Varma
Stanford University

Maximum Flow Formulation Identifies High-Confidence Noncoding Variants Associated With Autism Spectrum Disorder

Background: Machine learning approaches for predicting putative genetic variants from whole genome sequence (WGS) data are often limited by the presence of high-dimensional variant feature spaces, which lead to model instability and lack of generalizability. Here, we design a novel maximum flow formulation based on linkage disequilibrium (LD) to address this issue and extract a set of stable, high-confidence noncoding variants that are likely to be associated with Autism Spectrum Disorder (ASD).
Methods: We analyze 232,193 variants in simple repeat sequences (SRS), collected from WGS of 2182 children with ASD and 379 controls. We perform 5-fold cross-validation with a logistic regression classifier and extract variants assigned non-zero scores from each validation fold. Then, we assemble the five sets of variants in a maximum flow network subject to LD constraints.
Results: The maximum flow formulation allowed us to identify 50 stable regions (representing 55 variants). To determine if these SNPs can serve as a viable biomarker, we train a logistic regression classifier on this reduced feature set, which performed well on the test set (AUC-ROC=0.812) as well as on an independent dataset consisting of unique samples (AUC-ROC=0.922).
Discussion: These results support the involvement of repetitive regions in ASD, and our methodology allows for the creation of robust, interpretable, and scalable machine learning models that can identify predictive variants. To the best of our knowledge, such a method has never been used before for analysis of high-dimensional feature spaces. Our results help pave the way towards biomarker-based diagnosis methods for ASD.

POSTER ABSTRACT 25

Philip Shaw
NHGRI

Estimating the Heritability of the Brain's Structural Connectivity and Its Association With Changing Symptoms of Attention Deficit Hyperactivity Disorder

Background: Twin studies show that age-related change in symptoms of attention deficit hyperactivity disorder (ADHD) is heritable. However, the heritability of the development of the neural substrates that underlie ADHD is unknown. Here, we use longitudinal data to estimate the heritability of developmental change in the microstructural properties of white matter tracts and determine associations with change in ADHD symptoms.
Methods: 133 children from 51 nuclear families (34 with ADHD; 84 males), all with two assessments (age at baseline: 9.2 ± 3.1 years; follow up: 11 ±3.3) from which the annual rate of change in ADHD symptoms was determined. Diffusion tensor imaging (3T; 60 non collinear directions) estimated voxel level axial diffusivity (AD) and radial diffusivity (RD). Additive genetic heritability (h2r) of the annual rate of change in microstructural properties was calculated using Sequential Oligogenic Linkage Analysis Routines. Permutation tests corrected for multiple comparisons by assessing cluster-size significance over the white matter skeleton.
Results: Rates of change in microstructural properties were heritable in two voxel clusters within the left uncinate fasciculus (AD: h2r =.48±.09, p=0.004) and the forceps minor (RD: h2r =.52±.12, p=0.01). Improvement in inattentive symptoms was associated with AD change in the left uncinate (t=3.41, p= .003), and with RD change in forceps minor (t=2.05, p= .06). The uncinate cluster was also associated with improvements in hyperactivity/impulsivity (t=-3.39, p= .003).
Discussion: We demonstrate heritability in the development of microstructural properties of some white matter tracts, and find these properties are also associated with change in ADHD symptoms.

POSTER ABSTRACT 26

Jason Lambert
University of California Davis

In Vivo Deployment of a Massively Parallel Reporter Assay for the Validation of Disease-Relevant Enhancers Active in Postnatal Brain Development

Enhancers recruit tissue- and cell-type-specific transcription factors to drive specific expression patterns. The regulatory activity of enhancers is thought to produce and organize gene expression patterns producing the vast diversity of cell types that make up tissues and organs in animal development. Various means of assessing chromatin state have proven useful in predicting the presence of enhancers, but it remains expensive and labor intensive to functionally validate predicted enhancers in vivo. However, recent advances in massively parallel reporter assays (MPRAs) make possible the large-scale screening of enhancers. We present progress toward functionally validating a set of predicted human enhancer sequences which contain single nucleotide polymorphisms associated with various neuropsychiatric diseases including schizophrenia, bipolar disorder, autism, and epilepsy in the neonatal mouse brain via the MPRA approach using recombinant adeno-associated virus as an expression vector. We also show that an enhancer sequence validated by this screen drives reproducible expression of GFP in neurons in the mouse cerebral cortex during early postnatal development. Moving forward, this approach will allow us to validate and dissect the regulatory function of disease-relevant enhancers across multiple time-points and cell types in postnatal brain development.

POSTER ABSTRACT 27

Marci Rosenberg
University of California San Francisco

Noradrenergic Regulation of Grey-Matter Astrocyte Maturation

Synaptic dysfunction has been observed in many psychiatric disorders. Increasingly, grey-matter astrocytes are being recognized for their crucial synapse-supporting roles, including promoting synaptogenesis and recycling neurotransmitters. Our lab previously defined interleukin-33 (IL-33) as a molecular marker of grey-matter astrocytes. Transcriptomic analysis of IL-33-positive astrocytes identified multiple genes that encode different subclasses of adrenergic receptors as being significantly upregulated. This suggests that norepinephrine (NE) may be an important molecular cue guiding astrocyte progenitors towards a grey-matter astrocyte fate. By depleting noradrenergic signaling via postnatal toxin administration or by deleting adrenergic receptors in a genetic knockout, I observed an expansion of glial fibrillary acidic protein (GFAP)-positive astrocytes into the cortex. Moreover, sparse labeling of individual astrocytes showed an increase in GFAP signal in the astrocytes of animals that received a NE-depleting toxin. GFAP is an established marker of white-matter and reactive astrocytes, both of which are non-synapse-supporting astrocyte subclasses. These findings raise the possibility that release of NE from neurons promotes the maturation of astrocytes into grey-matter astrocytes that support synapses.

POSTER ABSTRACT 28

Amanda Welch
University of California San Diego, Department of Psychiatry

Dopamine D2 Receptor Overexpression in the Nucleus Accumbens Core Indirect Pathway Increases Activity-Based Anorexia Selectively in Female Mice

Anorexia nervosa (AN) is an eating disorder characterized by severe hypophagia, weight loss, and intense fear of weight gain. In the activity-based anorexia (ABA) paradigm, rodents exposed to running wheels and restricted food access exhibit extreme weight loss, hypophagia, and hyperactivity. Days of survival in the ABA paradigm provide a measure of ABA susceptibility, which models aspects of AN. Human imaging studies reported that recovered female AN patients show increased D2/D3R binding in the ventral striatum. Eight week old transgenic male and female Drd2-cre mice received infusions of an AAV separately expressing the long form of D2R and mVenus, or a control virus expressing only EGFP in a Cre-dependent fashion. Four weeks later, mice were tested in open field. Our results showed increased locomotor activity in mice overexpressing D2Rs ($P < .0001$). Mice were then singly housed with a running wheel with food and water ad lib during baseline. Bodyweight, food consumption, and wheel running measures were collected daily for 4 days. After the baseline period, food was available 7 hours each day. During the restriction phase, the same measures were collected daily, including days of survival. Our results showed that mice overexpressing D2Rs in ventral striatal indirect pathway neurons showed reduced survival in the ABA paradigm ($P < .05$) compared to controls. This effect was observed in female ($P < .05$), but not male mice. Our findings suggest that overexpression of D2Rs on ventral striatal indirect pathway neurons increase ABA behavior and may play a causal role in the development of AN.

POSTER ABSTRACT 29

Vipavee Niemsiri
Department of Psychiatry, University of California San Diego

Integrating the iPSC-Derived Neuron Transcriptome and GWAS-Boosting Data to Identify Genes and Networks Involved in Lithium Response in Bipolar Disorder

Background: Bipolar disorder (BD) is a severe psychiatric condition requiring long-term treatment. Lithium (Li) is the first-line mood stabilizer to which approximately, one-third of patients respond. However, identifying such patients a priori is difficult. The aim of our study was to determine potential genes and pathways involved in lithium response in BD.

Methods: We performed RNA-sequencing (RNA-seq) analysis in patient-derived induced pluripotent stem cells (iPSC) neurons and compared between BD Li-Responders (LR, n=6) and BD Li-Non-responders (NR, n=5) treated in vitro both with and without lithium. Subsequently, we integrated differentially expressed (DE) genes and the top 5% genome-wide association study (GWAS)-boosting (GWAB) genes (n=1120) into the network propagation analysis, followed by the pathway enrichment analysis.

Results: We identified 41 nominally significant DE genes ($p<0.05$, log2fold-change$\geq|1|$ with false discovery rate [FDR]≤ 0.20) in iPSC neurons in LR relative to NR regardless of in vitro lithium exposure. Seventy-three out of 1120 GWAB genes overlapped ($p=1.3E-09$) with the top 500 network derived genes proximal to the 41 DE genes. The pathway analysis in the 500 network genes revealed enrichment in 37 KEGG pathways (FDR<0.05), including focal adhesion, ECM-receptor interactions, and PI3K-Akt signaling.

Conclusions: GWAS power can be increased substantially by using RNA-seq data. The pathways we found enriched include several known to be modulated by lithium. Additionally, we found evidence for the involvement of focal adhesion, gap junction and ECM -receptor interaction pathways, to our knowledge, not previously implicated.

POSTER ABSTRACT 30

OPEN BOARD

POSTER ABSTRACT 31

Vivek Kumar
The Jackson Laboratory

Tmod2 is a Regulator of Cocaine Responses Through Control of Striatal and Cortical Excitability, and Drug-Induced Plasticity

Drugs of abuse induce neuroadaptations, including synaptic plasticity, that are critical for transition to addiction, and genes and pathways that regulate these neuroadaptations are potential therapeutic targets. Tropomodulin 2 (Tmod2) is an actin-regulating gene that plays an important role in synapse maturation and dendritic arborization and has been implicated in substance-abuse and intellectual disability in humans. Here we mine the KOMP2 data and find that Tmod2 knockout mice show emotionality phenotypes that are predictive of addiction vulnerability. Detailed addiction phenotyping showed that Tmod2 deletion does not affect the acute locomotor response to cocaine administration. However, sensitized locomotor responses are highly attenuated in these knockouts, indicating perturbed drug-induced plasticity. In addition, Tmod2 mutant animals do not self-administer cocaine indicating lack of hedonic responses to cocaine. Whole brain MR imaging shows differences in brain volume across multiple regions although transcriptomic experiments did not reveal perturbations in gene co-expression networks. Detailed electrophysiological characterization of Tmod2 KO neurons showed increased spontaneous firing rate of early postnatal and adult cortical and striatal neurons. Cocaine-induced synaptic plasticity that is critical for sensitization is either missing or reciprocal in Tmod2 KO nucleus accumbens shell medium spiny neurons, providing a mechanistic explanation of the cocaine response phenotypes. Combined, these data provide compelling evidence that Tmod2 is a major regulator of plasticity in the mesolimbic system and regulates the reinforcing and addictive properties of cocaine.

POSTER ABSTRACT 35

Siavash Fazel Darbandi
University of California San Francisco

TBR1 Dosage is Required for Cortical Neuronal Spine Maturation and Synaptogenesis

An understanding of how heterozygous loss-of-function mutations in autism spectrum disorder (ASD) risk genes, such as TBR1, contribute to ASD remains elusive.Tbr1 is a high confidence ASD gene encoding a transcription factor with distinct pre- and postnatal functions. Postnatally, Tbr1 conditional mutants (CKOs) and constitutive heterozygotes have immature dendritic spines and reduced synaptic density. Tbr1 regulates expression of several genes that underlie synaptic defects, including a kinesin (Kif1a) and a WNT signaling ligand (Wnt7b). Furthermore, Tbr1 mutant corticothalamic neurons have reduced thalamic axonal arborization. Promoting WNT-signaling, robustly rescues the synaptic and axonal defects, suggesting that this could have relevance for therapeutic approaches in some forms of ASD.

POSTER ABSTRACT 39

Kelley Paskov
Stanford University

Microdeletions on the X Chromosome Contribute to Autism Susceptibility in Males

Background: Autism spectrum disorder is a neurodevelopmental condition affecting 1 in 53 children, with a male-female ratio of 3:1. Despite careful study of the X-chromosome due to disorders such as Turner, Rett and Fragile-X syndromes that are comorbid and share phenotypic characteristics with autism, few X-chromosomal variants have been definitively linked to idiopathic autism. However, most studies have focused on single nucleotide variants, due to the expense and difficulty of sequencing and calling other types of variants, such as deletions. Autosomal deletions are known to play an important role in many forms of syndromic autism, suggesting that deletions on the X-chromosome may contribute to autism susceptibility and explain autism's sex bias.
Methods: We use whole-genome sequencing data from more than 800 multiplex autism families to identify inherited deletions on the X chromosome ranging from 0.1-100Kb in size. By using a hidden Markov model to capture familial relationships, we are able to identify deletions in this size range with high fidelity in both males and females.
Results: We show that autistic males are more likely to inherit X-chromosomal deletions than their neurotypical siblings. We further identify a deletion in Xp22.32 that is transmitted to autistic males significantly more often than would be expected by chance.
Conclusion: Sequencing nuclear families makes it possible to identify inherited X-chromosomal deletions, which would otherwise be difficult to detect. These deletions contribute to autism susceptibility and may explain why males are more susceptible to the disorder than females.

POSTER ABSTRACT 40

Rebecca Shafee
Harvard Medical School

Genetic Architecture of Phenome-Wide Latent Factors in the UK Biobank

Background: Large-scale biobanks and electronic health records can offer unprecedented insight into the genetic architecture of a wide range of traits in the general population. However, the high dimensionality of phenotypic data, with thousands of traits measured per individual, can make it difficult to interpret genetic results. Here we derive and genetically characterize latent phenotypic factors representative of the spectrum of traits measured in UK Biobank (UKB).
Methods: Beginning with 4203 traits, we identified individuals and traits with low missingness (i.e., ~10%) in order to limit association with survey structure. We performed exploratory factor analysis in this core dataset of 33,860 individuals and 730 items. We also performed a genome-wide association study (GWAS) of each factor and used LD score regression to estimate heritability and also to calculate the genetic correlation of the factors to psychiatric and somatic disorders studies outside of UKB.
Results: The final model includes 36 stable, interpretable factors accounting for ~30% of total phenotypic variance and spanning a wide range of physical (e.g., body size, general pain), behavioral (e.g., neuroticism, smoking), and lifestyle (e.g., education, urbanicity) dimensions.
In preliminary results, genetic correlations confirm expected associations (e.g., rg=0.58 for coronary artery disease and the heart attack factor) and also reveal a surprisingly broad, diffuse pattern of genetic correlation between common psychiatric disorders and phenotypic factors.
Conclusions: Overall, our results suggest that phenotypic factor analysis enhances interpretability, boosts power for heritability analyses, and can yield meaningful reduction in dimensionality to drive the next generation of genetic studies.

POSTER ABSTRACT 41

Vasiliki Karalis
University of California Berkeley

Raptor Downregulation Prevents TSC-related Phenotypes in Mice

Tuberous sclerosis complex (TSC) is a multi-system disease caused by disruption of the TSC1 or 2 genes. TSC is associated with significant neurological and psychiatric problems including epilepsy, intellectual disability, and autism. Since the TSC1/2 genes form a protein complex that negatively regulates mTOR complex 1 (mTORC1) activity, current pharmacological approaches use drugs such as rapamycin and its derivatives that suppress mTORC1 signaling. While these drugs have been successful in some aspects, rapalogues do not fully suppress all pathways downstream of mTORC1 and chronic use indirectly impairs mTOR complex 2 (mTORC2) signaling. Our goal is to identify alternative approaches to improve the biochemical and developmental changes associated with TSC1/2 loss in the brain. Specifically, we are testing whether genetic reduction of the obligatory mTORC1 component Raptor, can prevent mTORC1 hyperactivity in in vitro and in vivo models of TSC. Our results in mouse hippocampal cultures demonstrate that genetic reduction of Rptor effectively rebalances mTORC1 signaling in the context of Tsc1 loss with minimal disruption of mTORC2. In addition, Rptor deletion prevents neuronal morphologic malformations resulting from Tsc1 loss. In vivo, mice with forebrain-specific loss of Tsc1 die prematurely. We have shown that deletion of Rptor prevents premature mortality in the Tsc1 knock out mice in a gene dose-dependent manner and prevents some of the TSC related brain anatomical phenotypes. This work provides novel information regarding the relationships between TSC1/2, mTORC1, and mTORC2 in neurons and suggests an alternative therapeutic strategy for treating TSC and other mTOR-related disorders.

POSTER ABSTRACT 42

Leah Dorman
University of California San Francisco

Defining the Roles of Glia in Developmental Cortical Remodeling

Background: Neural circuit maturation requires ongoing structural remodeling of synapses, glial cells, and vasculature. We recently identified a role for innate immune signaling between astrocytes and microglia in synapse remodeling, raising the question of how these cell types are more generally involved in refining neuronal circuitry. Astrocytes and microglia are poised to respond to changing neuronal input during cortical development, but it is unclear if and how these glial cells are involved in coordinating large-scale synapse remodeling.
Methods: We used the somatosensory cortex as a model to study the glial response to developmental synaptic remodeling. Whisker cauterization before postnatal day four leads to stereotyped changes in the topographic cortical representation of the remaining whiskers, requiring substantial synaptic rearrangement. We unilaterally cauterized alternating rows of whisker follicles in perinatal mice and used a combination of histology and single-cell RNA sequencing to examine the effects on astrocytes and microglia.
Results: Whisker cauterization led to topographic rearrangement in the barrel cortex without substantial synaptic loss. Both microglia and astrocytes altered their morphology and gene expression profiles, but without evidence of 'inflammatory' changes. The astrocyte response included increases in protease expression, while a population of microglia showed increased expression of genes associated with purinergic sensing and motility.
Conclusion: Both astrocytes and microglia respond to local cortical remodeling. Future work will determine the functional impact of these genetic changes and may identify a glial remodeling response that reveals evidence of plasticity in less anatomically defined circuits where neuronal architecture is shaped by alterations in experience.

POSTER ABSTRACT 43

Tracy Warren
University of California Davis Center for Neuroscience

Validation of Active Enhancers Within the Disease-Associated CACNA1C Gene

Background: Characterizing disease-associated enhancers is critical to understanding the link between genetic risk loci, gene regulation, and psychiatric disease. Multiple studies identified a linkage disequilibrium (LD) block within intron one of CACNA1C as associated with psychiatric disorders. We previously tested 22 sequences that harbored SNPs from this LD block for enhancer activity in a large functional screen. The sequences included all linked SNPs from the LD block. Here we validated three positive enhancers identified in the screen and initiate in-depth characterization of these disease-relevant CACNA1C enhancers.
Methods: Enhancer activity was tested in human cell culture via reporter assays. In parallel, we used a novel rAAV GFP reporter assay to test activity of candidates in postnatal mouse brain. Following P0 rAAV transduction, we collected P7 brains and performed image analysis to assess GFP expression in brains transduced with predicted positive and negative enhancers.
Results: This analysis validates that functional screening in mouse brain is indeed capable of identifying sequences with enhancer activity. The predicted enhancer sequences drove reporter expression in vitro in cells and in vivo in postnatal mouse cortex.
Conclusion: We show that three identified elements located within CACNA1C are active enhancers. Our ongoing work aims to characterize enhancer-based modulation of CACNA1C expression and to investigate activity of disease-associated alleles. A better understanding of how such enhancers control gene expression across cell types and brain regions will be key in moving from association toward elucidating underlying mechanisms of neuropsychiatric disorders.

POSTER ABSTRACT 44

Jerika Barron
University of California San Francisco

Determining the Role of Type 2 Immunity in Synapse Development

Immune signaling plays physiologic roles in neural circuit development. Conversely, immune dysregulation has been implicated in neurodevelopmental disorders including Autism Spectrum Disorder (ASD) and schizophrenia. Microglia are brain-resident macrophages that contribute to neuronal synapse development and are the primary immune cells within the brain parenchyma. However, the immune signals that regulate microglial function are unknown. Here, we have identified a potential source of such signals. We find that immune cells known as innate lymphocytes are present in the meningeal lining of the brain during postnatal development. We have focused on a specific subset of these innate lymphocytes, known as 'type 2' (ILC2) due to their known roles in tissue remodeling and repair. Previous work showed that cytokines produced by these lymphocytes (IL-13) are required for normal cognition and behavior, however their roles in neurodevelopment are largely unknown.

To test the hypothesis that IL-13 signals to microglia to promote neural circuit remodeling during development, we performed gain and loss of function experiments in mice and quantified synapse numbers and microglial transcriptional responses. We find that loss of ILC2 or of the receptor for IL-13 leads to a reduction of inhibitory synapse numbers in the developing somatosensory cortex as measured by immunohistochemistry and confocal imaging. Conversely, exogenous IL-13 leads to a robust transcriptional response in microglia and increases inhibitory synapse numbers. Future work will specifically test the hypothesis that loss of IL-13 signaling to microglia increases Excitatory/Inhibitory balance in the cortex, a phenotype that has been associated with neurodevelopmental disorders including ASD.

POSTER ABSTRACT 45

Jae-Yoon Jung
Stanford University School of Medicine

Identifying LCL Artifacts in Whole Genome Sequencing

Background: Lymphoblastoid cell lines (LCLs) have been extensively used in expression and genotyping projects, as a continuous source of genomic DNA from immortalized B cells. It is known that LCLs show high genotype concordance with their parental peripheral blood (WB) samples especially in their early passage, but little has been reported about their characteristics in large-scale whole genome sequencing (WGS) projects, where passage information is usually not available.
Methods: We show that in WGS, LCL samples tend to have more rare variants than WB samples, which complicates rare variant analysis. We build a machine learning (ML) model that is able to differentiate reliable rare variants from suspected LCL artifacts, training our model using twin samples and testing performance on matched LCL/WB samples from the same individuals.
Results: After applying our ML model to filter outliers, we demonstrate that there is no significant difference in the number of rare variants between LCL and WB samples, and they are in the expected range reported from other studies.
Conclusion: Our ML model to estimate LCL artifacts is trained with a large number of samples, tested against one of the largest WGS data sets currently available, and easily applicable to any other WGS data set with LCL samples. We make the first trained version publicly available, and the generalized model will follow as a next step.

POSTER ABSTRACT 46

Phi Nguyen
University of California San Francisco

The IL-33-IL1RL1 Signaling Axis Coordinates Remodeling and Integration of Adult-Born Neurons in the Hippocampus

Learning, memory, and neural circuit plasticity require continual dendritic and synaptic remodeling, which are perturbed in psychiatric disorders. In the mammalian brain, the dentate gyrus of the hippocampus is one of the few germinal niches that undergoes neurogenesis and dynamic circuit remodeling in response to novel experience throughout adulthood. In order to survive and contribute to hippocampal function, adult-born neurons must properly integrate into the local circuitry. However, the mechanisms that underlie this process are poorly understood. Here we show that the IL-1 family cytokine IL-33, which signals through its cognate receptor IL1RL1, is a signaling molecule highly enriched in the dentate gyrus that is required to maintain dendritic spine density and survival of adult-born neurons, consistent with a role in circuit integration. Microglia have been shown to participate in modulating synaptic plasticity and our group has previously shown that IL-33-IL1RL1 signaling promotes microglia engulfment in other CNS regions. In the hippocampus, we also find that the IL-33 receptor IL1RL1 is predominately expressed by microglia and loss of this receptor disrupts microglia ramification and coverage area. We propose that IL-33-IL1RL1 signaling facilitates microglial-mediated synapse and dendritic remodeling in the hippocampus. Future work aims to elucidate the mechanisms and functional implications of this pathway in hippocampal circuit plasticity.