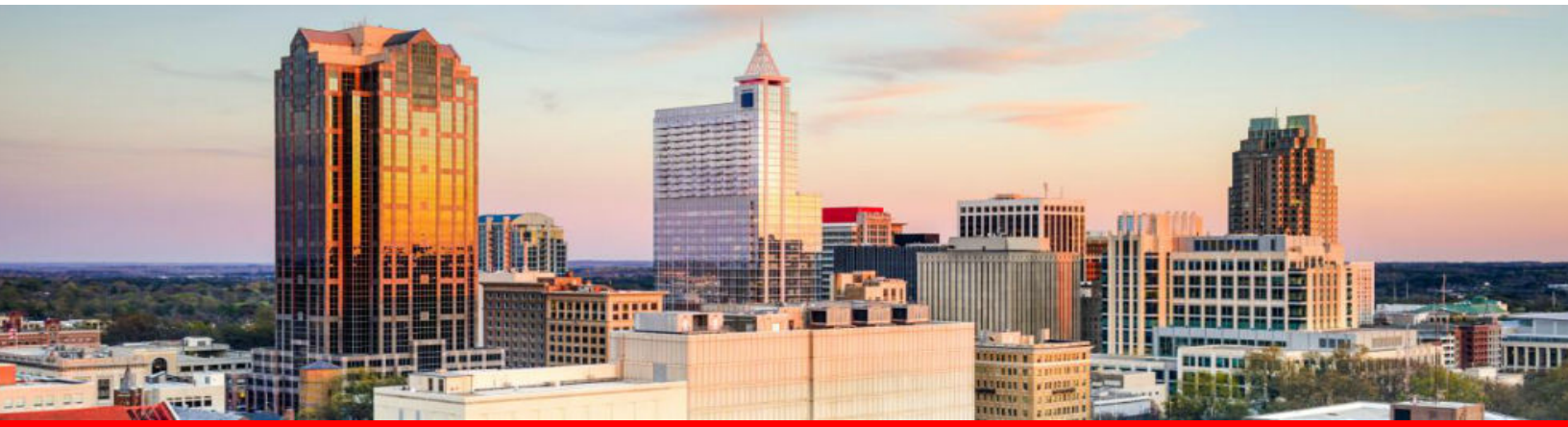
TRIANGLE CHAPTER
SOCIETY FOR NEUROSCIENCE

2nd Spring Neuroscience Conference



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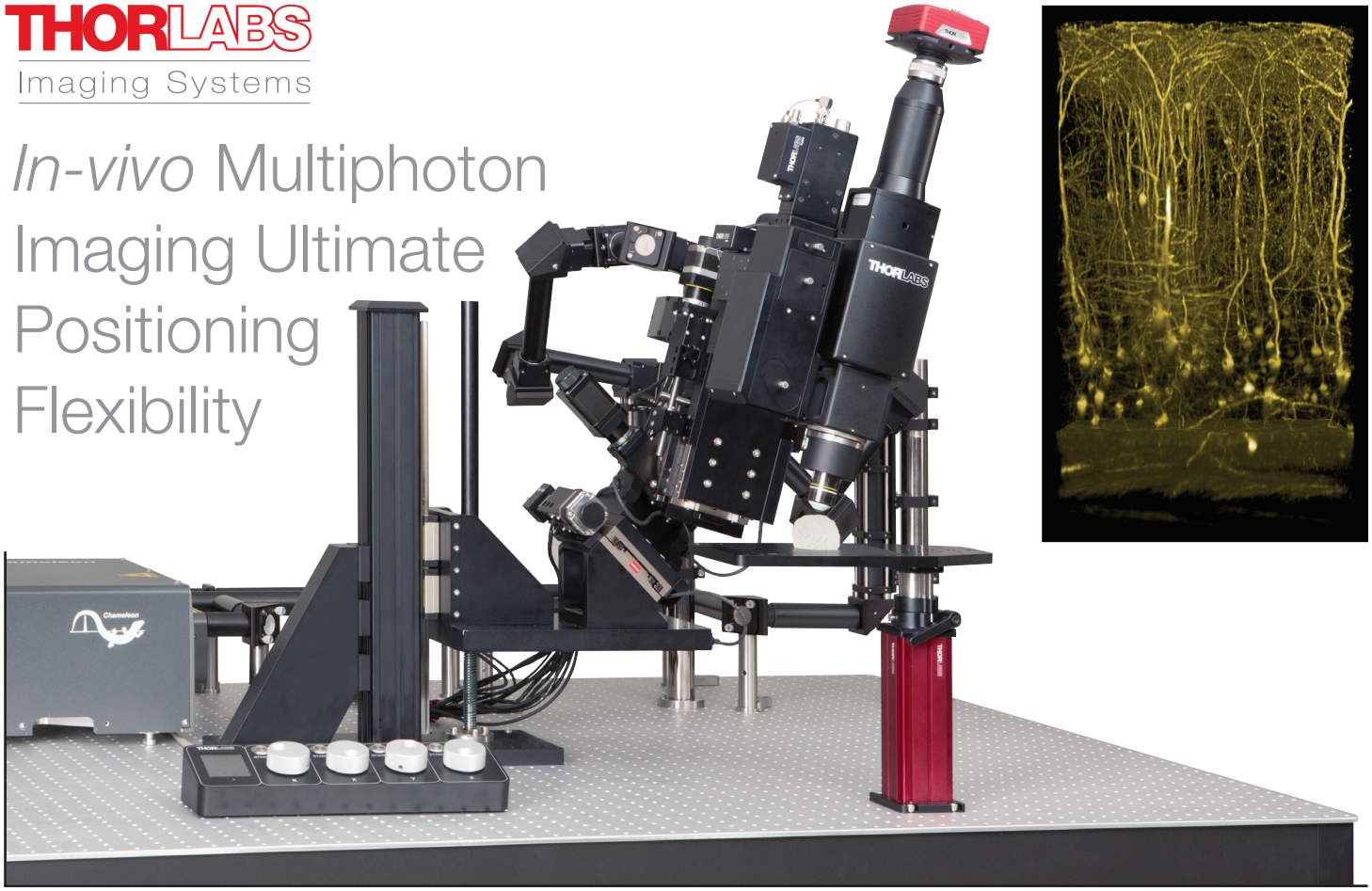

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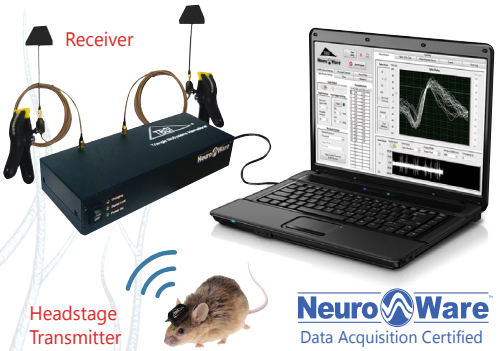

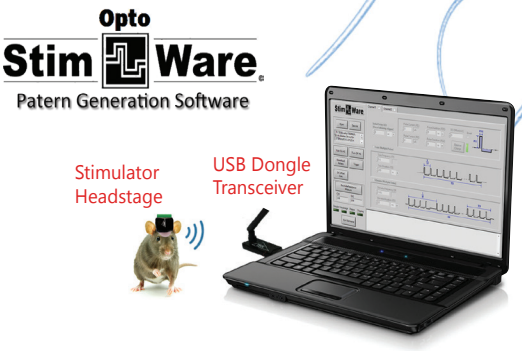

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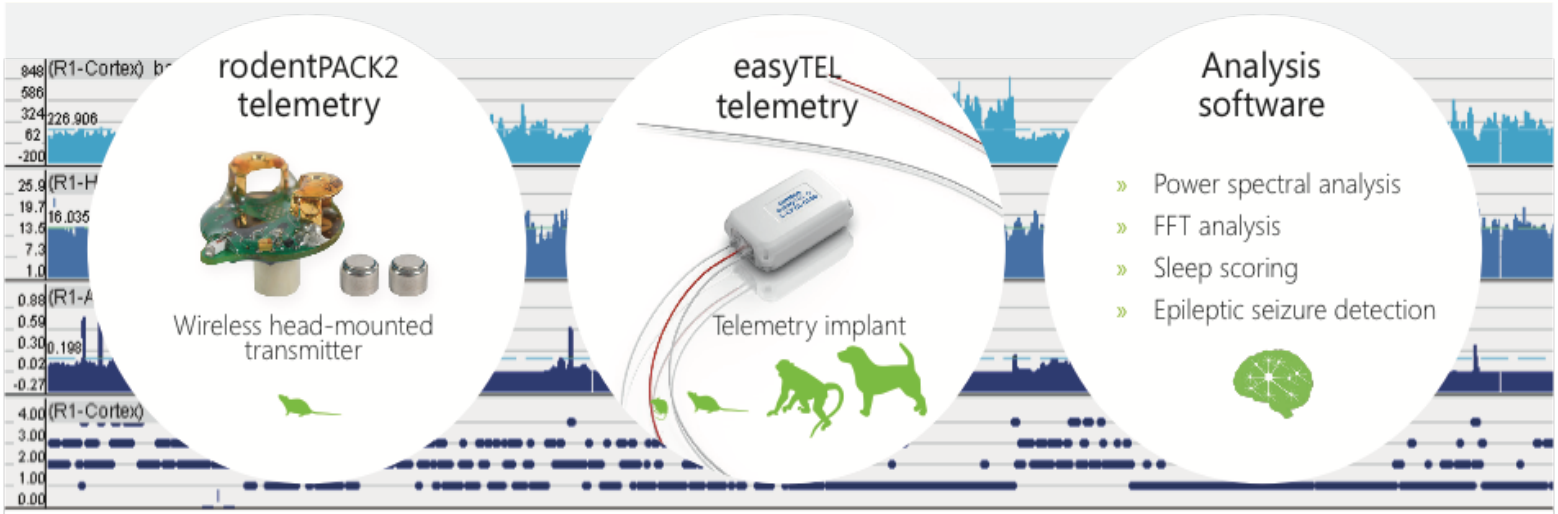

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TRIANGLE CHAPTER
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Program

1:00 – 2:30 Opening Remarks

Local Speaker Presentations:

Dr. Cagla Eroglu
Assistant Professor of Cell Biology
Duke University

Dr. Garret Stuber
Assistant Professor of Psychiatry
University of North Carolina at Chapel Hill

Dr. Wayne Silver
Professor of Biology
Wake Forest University

2:30 – 2:45 Coffee Break

2:45 – 3:45 Poster Presentations

3:45 – 4:45 Keynote Address:

Dr. Keith W. Kelley
Professor Emeritus of Immunophysiology
University of Illinois
Editor-in-Chief: *Brain, Behavior, and Immunity*

4:45 – 5:00 Poster Awards Ceremony

5:00 – 6:00 Reception

Speakers

Keith W. Kelley, PhD

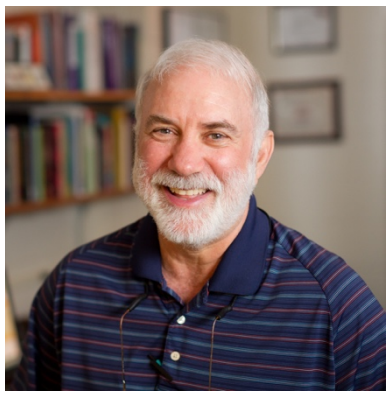
"Getting Nervous About Immunity"

Keith W. Kelley is a Professor Emeritus of Immunophysiology at the University of Illinois. Dr. Kelley earned his Ph.D. in 1976, a time when the immune system was considered only to protect against infectious diseases. He helped reshape that view by bringing physiology to immunology. It is now accepted that there is an active dialogue between the immune system and brain, and these discoveries are improving human and animal health.

Professor Kelley has been honored with 10 university and national awards, published more than 250 peer-reviewed scientific papers as well as 70 book chapters, is well cited with an h-index of 68, been funded for 50 years as the Principal Investigator of NIH grants and served on four dozen NIH study sections. He is currently a Full Member of the Neuroendocrinology, Neuroimmunology, Rhythms and Sleep Study Section. Dr. Kelley is a Past-President and Secretary-Treasurer of the PsychoNeuroImmunology Research Society. He has been Editor-in-Chief of *Brain, Behavior, and Immunity* since 2003. The latest impact factor of the journal is 5.9, ranking it in the top 14% of all immunology journals and 11% of neuroscience journals.

Cagla Eroglu, PhD

Garret Stuber, PhD



Wayne Silver, PhD

Remarks

Welcome to the 2nd Annual Spring Neuroscience Conference!

Colleagues and Friends,

It is with great pleasure and honor that I welcome you all to our second annual Spring Neuroscience Conference. Our chapter has grown significantly since it was reinstated two years ago, after many years of dormancy; we owe this tremendous success to our many active and motivated members and to the vibrant Triangle neuroscience community. Fostering collaborations within our community and promoting neuroscience to the general public have been our top priorities since our reinstatement, and I am pleased to report that our chapter has been successful in achieving these goals.

Some examples of our activities over the past year include: the successful scientist exchange program that we developed in collaboration with Wake Forest University; fruitful meetings with congressman David Price regarding NIH funding; co-sponsorship of the inaugural Dr. TK Li Distinguished Lectureship at Duke University; facilitation and partnership with numerous brain awareness week events in both 2015 and 2016; numerous lectures and workshops for local schools throughout the community; the inaugural Spring Neuroscience Conference in 2015; and, of course, today's second annual Spring Neuroscience Conference. As we move forward, we hope to provide more community outreach programs to engage a more diverse audience. I am confident that with your active participation, we will achieve this goal.

This year, we have invited four distinguished neuroscientists to speak at our meeting, including three from our own backyard. It is with great honor that we present lectures by Dr. Cagla Eroglu of Duke University, Dr. Garret Stuber of UNC-Chapel Hill, Dr. Wayne Silver of Wake Forest University. Additionally, we are pleased to introduce Dr. Keith Kelley of the University of Illinois as our keynote speaker.

I want to thank you all for joining us today. As I reach the end of my term as president of Triangle SfN, I would like to thank my wonderful team and chapter members for their enthusiasm and tireless efforts. I am confident that our new president, Dr. Patricia Jensen, will work hard to continue to grow our chapter. She has already shown that she is full of great ideas; I wish her the best in continuing to further the goals of our chapter. I strongly believe that with your active involvement, we can collectively achieve our objectives and further an appreciation for the importance of neuroscience research here in North Carolina. We rely on your help in this important journey.

Amir H. Rezvani, Ph.D.
President

Abstracts

Poster Number: 01

Differential Effects Of Tobacco Smoke Containing Either High Or Low Levels Of Nicotine During Adolescence On The Central Cholinergic System In Mice

Y Abreu-Villaca, A Nunes-Freitas, AC Dutra-Tavares, M Correa-Santos, D Paes-Branco, CC Filgueiras, AC Manhaes, A Ribeiro-Carvalho
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The central cholinergic system is the primary site of action of nicotine; however, other components of tobacco smoke may either directly cause some effects of smoking or interfere on nicotine actions on this system. In the present study, we investigated the effects of exposure of adolescent male and female mice to tobacco smoke containing either high or low levels of nicotine on the central cholinergic system and the effects associated with cessation of exposure. From postnatal day (PN) 30 to 45, Swiss mice were exposed to tobacco smoke (whole body exposure, 8h/day, 7days/week) generated from 2R1F (HighNic group: 1.74mg nicotine/cigarette) or 4A1 (LowNic group: 0.14mg nicotine/cigarette) research cigarettes, whereas control mice were exposed to ambient air. Cholinergic biomarkers were assessed in the cerebral cortex and midbrain by the end of exposure (PN45), at short- (PN50) and longterm (PN75) deprivation. In the cortex, nicotinic cholinergic receptor upregulation was observed with either type of cigarette. In the midbrain, upregulation was detected only in HighNic mice and remained significant in females at short-term deprivation. The high-affinity choline transporter was reduced in the cortex: of HighNic mice by the end of exposure, of both HighNic and LowNic females at short-term deprivation, and of LowNic mice at long-term deprivation. These decrements were separable from effects on choline acetyltransferase and acetylcholinesterase activities, suggesting cholinergic synaptic impairment. The current set of results extends previous evidence of deleterious effects of tobacco smoke exposure during adolescence, suggests that the sole reduction of nicotine yield in tobacco products does not spare the central cholinergic system, and paves the way for future studies to fully characterize the compounds responsible for these effects.

Poster Number: 02

Git/Pix As A Rac Effector

U Ahmed, R Premont
Department of Gastroenterology; Duke University; Durham, NC 27710

Small GTP-binding proteins (GTPases) are important regulators of neuronal functions, including motility, differentiation, and signaling. Mutations of these proteins can lead to a variety of pathological conditions, including neurodegenerative disorders. The Rho family of GTPases is important in regulating the dynamic cytoskeleton that allow for neuronal and synaptic functions. GIT/PIX, a protein complex that coordinates a number of signaling events, is a well-known upstream regulator of the Rac1 small GTPase, a member of the Rac-like branch of the Rho family of GTPases, but has also been reported to function upstream of GIT/PIX. The central hypothesis of this project is that all eight members of the Rac-like subfamily of the Rho GTPases can bind to and regulate GIT/PIX as an effector protein, and also are activated by GIT/PIX. We will investigate how binding of active Rac proteins to GIT/PIX alters association of GIT/PX binding partners, including paxillin and PAK3. Uncovering the protein interactions that surround the regulation of Rac-like proteins will help further understanding of how to develop effective therapies for neurodegenerative disorders

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Poster Number: 03

Chemo-Genetic Activation of Hippocampal Area CA2 Neurons Increases Gamma Oscillations in Hippocampus and Prefrontal Cortex

G. Alexander, L. Brown, S. Farris, C. Pantazis, D. Lustberg, B. Gloss, N. Plummer, P. Jensen, S. Dudek
Neurobiology Laboratory, NIEHS, RTP, NC, 27709

Gamma oscillations (30-120Hz) occur widely throughout the hippocampal formation where they are believed to play a critical role in the encoding and retrieval of declarative memories by coordinating the activation of local neuronal populations into transient cell assemblies. Gamma oscillations have warranted considerable attention in schizophrenia because individuals diagnosed with the disease display abnormalities in this frequency band during a variety of perceptual and cognitive tasks. Although relatively understudied, the CA2 field of the hippocampus is becoming increasingly recognized as a socio-cognitive hub for processing memories containing socially relevant information. Because impairments in social cognition are a common symptom associated with schizophrenia, and because CA2 is unique within the hippocampus in its susceptibility to loss of interneurons in schizophrenia, abnormal gamma activity in CA2 emerges as a candidate mechanism for linking the known cellular and network pathologies with the sociocognitive impairments observed in individuals with schizophrenia. To address the relationship between CA2 neuronal activity and gamma oscillations in hippocampus and prefrontal cortex, we infused adeno-associated viruses coding for a cre-

dependent excitatory DREADD (Designer Receptors Exclusively Activated by Designer Drugs) into hippocampi of mice that express cre recombinase selectively in CA2 pyramidal cells under the control of the Amigo2 gene. We then implanted electrode arrays to monitor activity of hippocampal and prefrontal cortical neurons before and following administration of the DREADD ligand, Clozapine-N-oxide (CNO), while animals freely behaved in an open field. We found that CNO increased firing of CA2 pyramidal neurons and dose-dependently increased power of local field potentials in the gamma frequency range in both and hippocampus and prefrontal cortex. At the level of the healthy brain, these findings demonstrate that activation of CA2 neurons is sufficient to induce gamma oscillations not only within the hippocampus, but also in the PFC, suggesting that the CA2 circuitry may play into distributed neuronal networks beyond its target, CA1. In the diseased brain, these findings support the idea that the loss of interneurons from CA2 may underlie, at least in part, the abnormal gamma oscillations observed in schizophrenic patients and provide a mechanistic link to the sociocognitive impairments observed in patients with schizophrenia

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Poster Number: 04

A Candidate Screen To Identify Novel Regulators Of Astrocyte Development And Astrocyte-Synapse Interaction

K Baldwin, J Stogsdill, C Eroglu
Department of Cell Biology, Duke University, Durham, NC 27710

Dysfunctional synaptic connectivity is thought to be the underlying cause of many neurological disorders, including autism, schizophrenia, and various forms of intellectual disability. Synaptic connections in the brain are a three part entity, consisting of the presynaptic axon, the postsynaptic dendrite, and the ensheathing astrocyte process. Research from our laboratory and others revealed that astrocytes structurally and functionally interact with synapses to actively participate in brain function. Disruption of astrocyte-synapse interactions is a major pathological mechanism seen in many neurological disorders, yet whether astrocyte dysfunction contributes to synaptic pathologies in neurological disorders is largely unknown. Previous studies have demonstrated that contact with astrocytes is critical for the ability of neurons to form and maintain synapses. Thus, defects in astrocyte development that impair the ability of astrocytes to develop extensive processes to ensheathe synapses are liable to impair the proper formation and function of synaptic connections By cross-referencing disease-linked genes to a database of cell-specific gene expression analyses in the murine CNS, we found that a number of these genes encode cell surface proteins that are highly expressed and significantly enriched in astrocytes compared to neurons. However, the function of these genes in astrocytes, and how disruption of their astrocytic function can contribute to neurological disorders, is not known. We hypothesized that many of these astrocyte-enriched disease-linked genes play critical roles in astrocytes to regulate astrocyte development and astrocyte-synapse interactions, and that disruption of their functions in astrocytes critically contributes to synaptic pathologies associated with neurological disorders. To test this hypothesis, we employed an astrocyte and cortical neuron co-culture system to screen carefully selected candidate genes for their role in astrocyte development and astrocyte-synapse interaction in vitro. Using this setup, we found that depletion of select candidate genes significantly impaired the ability of astrocytes to establish a complex morphology. Ongoing studies are focused on determining the in vivo astrocytic function of these candidate genes using a concurrent labeling and genetic modification strategy. Collectively, these experiments will provide novel insights into the role of astrocyte dysfunction in neurological disorders, and reveal specific molecular mechanisms that facilitate disease pathogenesis.

Poster Number: 05

Development And Validation Of An Electronic Cigarette Aerosol Exposure System For Rodents

D Barrus, T Lefever, R Cortes
RTI International, Research Triangle Park, NC, 27709

In recent years, electronic cigarette use has increased dramatically, with global sales estimated to be over $7 billion in 2015. Among adolescents, electronic cigarettes have recently outpaced tobacco cigarettes in popularity. The broad appeal of these devices stems from their perceived ability to deliver nicotine without the health risks associated with smoking. Increasingly, electronic cigarettes are also often used as a method of administering drugs of abuse (e.g., marijuana extracts, synthetic cannabinoids, stimulants). However, the majority of pre-clinical research into substances that are administered using electronic cigarettes rely on parenteral routes of administration that may not accurately reflect the physiological and behavioral effects of the aerosol products to which electronic cigarette users are actually exposed. In order to address this issue, we designed a system that delivers aerosol generated from a commercially available electronic cigarette to a mouse enclosed in a chamber. In Experiment 1, we administered vehicle or nicotine to adult ICR mice via aerosol (5 min exposure) or injection (SC) and measured subsequent effects on body temperature and locomotor activity. Three concentrations/doses of nicotine were tested. In Experiment 2, we pretreated all mice with mecamylamine (SC), a nicotinic acetylcholine receptor antagonist, and repeated the procedure of Experiment 1 in order to ensure that our system was delivering aerosol containing nicotine. Our system successfully produced dose-dependent effects typical of nicotine and these effects were antagonized by mecamylamine. Aerosol and parenteral administration of

nicotine resulted in a similar profile of effects, though there were some differences in time course. These results validate the use of our system to further assess the behavioral, toxicological, and pharmacokinetic effects of nicotine administered via electronic cigarettes. Further pre-clinical research using this system will provide insights into the effects of electronic cigarette use that should be more translational to actual human use than results found using parenteral administration

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Poster Number: 06

Diacylglycerol Kinase Iota (Dgkι) Regulates Pain Signaling In Peripheral Nociceptive Neurons

V Bartsch, M Zylka
Cell Biology and Physiology Department, University of North Carolina at Chapel Hill, Chapel Hill, NC 27514

Chronic pain drastically impacts daily function, often hindering work performance and severely impairing quality of life. Current treatments efficacy for long-term use. Characterizing the molecular mechanisms by which painful stimuli are processed may reveal candidate molecules to target with novel pharmacological treatments. Pain-sensing neurons (nociceptors) whose cell bodies lie in the dorsal root ganglia (DRG) detect noxious stimuli in peripheral tissues and transmit the signal to the spinal cord, then to the brain. Inflammatory mediators released after injury both activate nociceptors, leading to the perception of pain, and sensitize nociceptors, causing enhanced pain responses upon further stimulation. The inflammatory molecules that contribute to this sensitization activate several receptor classes, including Gαq-protein-coupled receptors (Gαq-GPCRs), receptor tyrosine kinases (RTKs), and transient receptor potential (TRP) channels. Gαq-GPCR and RTK activation yields diacylglycerol (DAG) production. DAG activates protein kinase C (PKC), which phosphorylates receptors to alter their activity. Phosphorylation by PKC desensitizes GPCRs and RTKs, but sensitizes TRP channels. Phosphorylation of DAG by diacylglycerol kinase (DGK) can alter DAG levels and subsequent activation of PKC. Therefore, DGK potentially regulates PKC-mediated modulation of receptor activity. We have identified the DGK isoforms that are most highly expressed in nociceptive DRG neurons of mice and humans. Of these, we have found that the iota isoform, DGK iota (DGKι), has the greatest kinase activity. Our data show that DGKι enhances Gαq-GPCR-induced calcium activity, and work by others demonstrates a similar effect on RTK signaling. We hypothesize that DGKι regulates GPCR and RTK signaling in peripheral nociceptors and modulates pain sensitization. We are investigating responses to nociceptive stimuli that signal through the GPCRs and TRP channels expressed in the DGKι+ DRG neurons. Our pilot data using DRG neurons cultured from our DGKι knockout mouse suggest that DGKι loss diminishes Gαq-GPCR-induced calcium responses without affecting TRP-mediated calcium influx. Furthermore, we have begun to examine pain sensitivity in response to noxious mechanical and thermal stimuli in DGKι knockout mice, assessing how perturbing specific pathways can manifest in different behavioral responses. In vitro signaling experiments coupled with in vivo pain-sensing behavioral assays will help us characterize the role of DGKι in nociception and discover the potential of this kinase as a target for analgesic therapies.

Poster Number: 07

White Matter Integrity Is Related To Cognitive Ability In Early Life

J Bullins, B Goldman, S Short, R Knickmeyer, M Styner, J Gilmore
Psychiatry; University of North Carolina; Chapel Hill, NC 27599

Background: Mounting evidence reveals white matter integrity as an indicator of cognitive ability in children and adults. However, little is known about how white matter fibers mature to support cognitive development in early life. In the present study we explored relationships between in-vivo white matter microstructural properties and cognitive ability in the first year of life.
Methods: Tract-based diffusion properties (FA, MD, RD, AD) were computed from diffusion tensor imaging (DTI) scans of 214 healthy 1-year-olds. Cognitive measures from the same subjects were collected using the Mullen Scales of Early Learning (MSEL). Functional mixed effects models assessed global and local diffusion properties in relation to MSEL scores.
Results: More mature microstructural properties in the left and right inferior longitudinal fasciculi (ILF) (greater FA, $p=0.01$, $p=0.02$) and corticothalamic tracts (CT) (lower MD, RD, and AD, $p \leq 0.05$) were related to higher general cognitive ability in 1-year-olds. FA in the right uncinate (UNC) was also related to cognitive ability ($p = 0.02$). A particular regional of significance in RD was located in the left CT tract passing through the internal capsule. When controlling for total brain size and cognitive ability, females exhibited generally higher FA in the UNC, ILF, and CT tracts ($p \leq 0.05$).
Conclusions: More mature tract properties along major white matter bundles related to cognition in adults were associated with greater cognitive ability in 1-year-olds. These tracts also exhibited gender differences. Results from this study suggest that white matter integrity in early life is important for determining cognitive ability

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Poster Number: 08

Enabling Real-Time Measurements Of Opioid Neuropeptides - Key Molecules Underlying Pleasure And Pain

Sarah Elizabeth Calhoun

Pain management is one of the oldest problems in medicine. Current treatments generally include opiate drugs which bind to opioid receptors in the brain; however, these neuronal targets are also involved in hedonic and motivational aspects of reward processing. A precise understanding of how opioid neuropeptides underlie pleasure and pain is lacking largely due to an inability to monitor dynamic fluctuations of these endogenous species in vivo. We are developing electrochemical technologies and implementing protocols for measuring the release of endogenous opioid peptides in brain tissue with fast scan cyclic voltammetry. This novel approach offers high spatial and temporal resolution with sufficient sensitivity and selectivity for measuring a specific opioid neuropeptide, methionine-enkephalin (mENK), in brain tissue. We have exploited every fundamental aspect of our waveform to optimize application rate, holding potential, accumulation potential, scan rate and switching potential allowing us to overcome a multitude of challenges associated with the quantitation of mENK in vivo. This optimized tool promises to advance the understanding of the role(s) mENK plays in numerous neuronal circuits and could be instrumental in revealing a therapeutic potential for a variety of behavioral and neuronal disease states.

Poster Number: 09

Gut Microbiome Associated With Brain Structure In Human Infants

A Carlson, K Xia, A Azcarate-Peril, M Styner, J Gilmore, R Knickmeyer Santelli
Neurobiology Curriculum, University of North Carolina at Chapel Hill, Chapel Hill, NC 27599

Background: Commensal gut microbiota has vital influence on health and development. Bidirectional signaling between the gastrointestinal tract and brain is influenced by the microbiome. Differences in microbe diversity are associated with temperament in children and impacts anxious behavior in rodents. Here we explored the association of gut microbial composition with brain tissue volumes and cognitive data during a critical period of neurodevelopment and gut colonization.
Methods: 89 typically developing infants were recruited at 1 year of age. Structural magnetic resonance brain scans were acquired on Siemens Allegra or Trio 3T scanners during unsedated sleep. Infant fecal samples were used to analyze the gut microbiome through Illumina sequencing of the V1-V2 region of bacterial 16s rDNA. Sequencing data analysis was prepared with QIIME. We used distance metrics and cluster scoring methods to identify genus level enterotypes within the group. Finally, ANOVA was used to test for differences in global brain tissue, 90-region grey matter volumes, and Mullen Scales of Early Learning outcomes between enterotypes.
Results: There was moderate support for clustering subjects into three enterotypes in this sample based on distance metrics and cluster scoring. Each cluster is characterized by the presence or absence of bacterial genera. Many genera showed variation across the 3 clusters including Bacteroides, Veillonella, and Faecalibacterium. There were no significant differences in global tissue volumes or cognitive scores, but several grey matter regions were found to be significantly different between clusters.
Conclusions: This cross-sectional study shows an association between the gut microbiome and brain structure during a foundational and vulnerable period in neurodevelopment. Future research into microbiome modulation of the gut-brain axis provides a potentially non-invasive way to intervene in abnormal developmental trajectories.

Poster Number: 10

Perineuronal Nets Suppress Plasticity Of Excitatory Synapses In Ca2 Pyramidal Neurons

Kelly Carstens1,2, Daniel Lustberg3, and Richard J. Weinberg1,2, Serena M. Dudek1,2,3
1Curriculum in Neurobiology, 2UNC Neuroscience Center, University of North Carolina, Chapel Hill, NC 27599, USA, 3Neurobiology Laboratory, National Institute of Environmental Health Sciences, NIH, Research Triangle Park, NC 27709

Perineuronal nets (PNNs) are a specialized form of extracellular matrix proteins that ensheath specific neuron cell types in the brain and spinal cord. The matrix concentrates predominantly around inhibitory interneurons and has been functionally implicated in inhibiting synaptic plasticity during postnatal development. Here we establish that PNN-associated immunofluorescence (wisteria floribunda agglutinin; WFA) labels mouse pyramidal neurons in CA2 and appears to be in proximity of excitatory synapses in CA2 stratum radiatum. Electron micrographs reveal WFA stain localized to the membrane of CA2 pyramidal cell somata and around dendritic spines. mRNA transcripts for the major PNN component aggrecan were also highly enriched in CA2. We found that rearing animals in an enriched environment increased staining for WFA nearly two-fold in CA2 at juvenile ages compared to control animals reared in standard housing. CA2 pyramidal neurons differ from other pyramidal neurons in hippocampus because they are resistant to the induction of synaptic long-term potentiation (LTP; Zhao 2007). We tested whether degradation of PNNs with chondroitinase (ChABC) permitted induction of LTP in CA2 neurons. We found that LTP could be induced in CA2 neurons after acute degradation of PNNs, matching the level of potentiation normally observed in CA1. Finally, we found that PNNs developed precociously in CA2 of a mouse model of Rett Syndrome (MeCP2 KO mice). Together these data indicate that PNNs around a population of excitatory hippocampal neurons regulate synaptic plasticity at excitatory synapses

and are regulated by experience during postnatal development.

Poster Number: 11

Intrathecal Bone Marrow Stromal Cells Inhibit Neuropathic Pain Via Tgf-Beta Secretion And Target Dorsal Root Ganglia Via Cxcl12

Gang Chen, Chul-Kyu Park, Rou-Gang Xie,Ru-Rong Ji
Department of Anesthesiology and Neurobiology, Duke University Medical Center, Durham, NC 27710

Neuropathic pain remains a pressing clinical problem and there are still no drugs that can treat neuropathic pain in a complete and definitive way. Bone marrow stromal cells (BMSCs) are a population of progenitor cells of mesodermal origin and present in the bone marrow of adults, emerging as a major source for cell-based therapies for clinical applications. BMSCs were originally conceived as stem/progenitor cells to rebuild diseased or damaged tissues. However, systemically infused BMSCs have been shown to exert therapeutic effects through the release of cytokines/trophic factors that act on local, or perhaps distant, target tissues. Through this paracrine modulation, it has been discovered that BMSCs are potent modulators of immune responses in humans and animals. Here we report that a local injection of BMSCs, via intrathecal route following lumbar puncture, can prevent and reverse neuropathic pain symptoms (allodynia and hyperalgesia) in mice for several weeks following nerve injury (chronic constriction injury, CCI). Intratehcal BMSCs also reduced CCI-induced ongoing pain as measured by conditioned place preference. Furthermore, this BMSCs treatment protected dorsal root ganglion (DRG) neurons from nerve injury and inhibited neuroinflammation in DRGs and spinal cords. Interestingly, BMSCs secreted TGF-β1 to CSF, and the analgesic effect of BMSCs was reversed by neutralization of TGF-β1 but not IL-10. Conversely, intrathecal administration of TGF-β1 potently inhibited neuropathic pain. TGF-β1 is a powerful neuromodulator and rapidly (within minutes) suppressed CCI-evoked spinal synaptic plasticity and DRG neuronal hyperexcitability via TGF-β receptor-1-mediated non-canonical signaling. CCI also upregulated CXCL12 in lumbar L4-L6 DRGs, and this up-regulation caused migration of intrathecally injected BMSCs to L4-L6 DRGs through CXCR4 expressed on BMSCs. These migrated BMSCs survived in DRGs for more than two months and eventually disappeared. Finally, intrathecal BMSCs also effectively reduced neuropathic pain after spared nerve injury. Our findings support a paracrine mechanism by which intrathecal BMSCs target the CXCL12-producing DRGs via to elicit neuroprotection and sustained neuropathic pain relief via TGF-β1 secretion.

Poster Number: 12

Deciphering The Role Of Norepinephrine In Brain Development

I Evsyukova, N Plummer, Y-W Chen, and P Jensen
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Noradrenergic neurons of the locus coeruleus (LC) project across the entire neuraxis and are the major source of norepinephrine to the neocortex. Given the early differentiation of LC neurons, their ability to synthesize and store norepinephrine long before their target regions mature, as well as the presence of noradrenergic afferents among the earliest inputs to the developing neocortex, we hypothesize that these neurons play a crucial role in neocortical development. To test this hypothesis, we generated a fluorescently tagged conditional knockout allele of dopamine β-hydroxylase (Dbh), a gene that encodes the enzyme that converts dopamine to norepinephrine. To gain access to LC neurons, we took advantage of the fact that these neurons share a history of En1 and Dbh expression and crossed Dbh cKO mice with En1Cre. We found that selective disruption of norepinephrine in the LC resulted in deficits in growth, neocortical development, and behavior. Neonatal mutants were mildly runted (80-90% of their littermate weight), suggesting that LC norepinephrine regulates metabolic state. Metabolic profiling experiments are now underway to determine whether levels of hormones that regulate lactation, feeding, and growth are altered in the mutants. We also observed several structural abnormalities in the developing brains of the mutants, including smaller overall brain size, thinner cortical wall, and smaller ventricles. We are now performing detailed characterization of neocortical layering and arealization patterns, and assessing survival and proliferation of neocortical progenitor cells to determine the mechanism by which selective norepinephrine disruption leads to these structural abnormalities. Pilot behavioral experiments (open field, elevated plus maze, light-dark box, and marble burying assay) on a cohort of adult mutant and littermate control mice showed no changes in the anxiety levels of the mutants. However, locomotion of the mutant animals was reduced in the light-dark box and open field tests, and the mutants spent more time in the center of the elevated plus maze, suggesting that disrupting norepinephrine production in the LC may affect locomotor and/or cognitive abilities. Taken together, our findings suggest that disrupted norepinephrine signaling in early development leads to structural changes in the neocortex and may result in impaired behavior in adulthood.

Poster Number: 13

Cross-Hemispheric Dopamine Projections Have Functional Significance

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Dopamine signaling occurs on a subsecond timescale, and its dysregulation is implicated in pathologies ranging from drug addiction to Parkinson's disease. Anatomical evidence suggests some dopamine neurons have cross-hemispheric projections, but the significance of these projections is unknown. Here, we show unprecedented interhemispheric communication in the midbrain dopamine system of awake and anesthetized rats. In the anesthetized preparation, optogenetic and electrical stimulation of dopamine cells elicited physiologically relevant dopamine release in the contralateral striatum. Contralateral release differed between dorsal and ventral striatum due to differential regulation by D2-like receptors. In the freely moving animal, simultaneous bilateral measurements revealed dopamine release synchronizes between hemispheres and intact, contralateral projections can release dopamine in the midbrain of 6-hydroxydopamine lesioned rats. These experiments are the first to show cross-hemispheric synchronicity in dopamine signaling and support a functional role for contralateral projections. In addition, the data reveal that psychostimulants such as amphetamine promote the coupling of dopamine transients between hemispheres.

Poster Number: 14

A Method To Visualize Visceral Sensing In Vivo

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Mood, anxiety, and appetite are a few behaviors modulated by gastrointestinal chemosensation. Although the mechanisms behind visceral processing are poorly understood, the sensory cells are known - they are called enteroendocrine cells. So far, the study of sensory processing by enteroendocrine cells have relied upon indirect methods, such as fixed tissue, cell lines, and calcium loading dyes. Here, we developed a method to study the calcium activity of enteroendocrine cells in vivo.
The method has three components: 1) a transgenic mouse in which enteroendocrine cells that secrete cholecystokinin (Cck) also express the fluorescent calcium sensing protein, GCaMP6s; 2) an abdominal window with a 3D printed scaffold to maintain intestine position against abdominal window and allow for chronic imaging of specific segment of intestine; and 3) a two-photon intra-vital imaging protocol to define oscillating calcium activity in enteroendocrine cells.
We implanted the abdominal window and scaffold into a CckGCaMP6s mouse and used two-photon microscopy to image as deep as 2mm into the gut wall. Basal calcium activity was recorded in enteroendocrine cells of the small intestine for 10 consecutive minutes at 1.16 frames per second. Our data show that enteroendocrine cells have intrinsic calcium oscillations at a rate of 6.0 ± 1.2 oscillations per minute (n=2). These results suggest that enteroendocrine cells are continuously active, even in the absence of nutrient stimuli, and open a new method to study of gastrointestinal chemosensation in vivo.

Poster Number: 15

Differences In Performance On The Antisaccade Task In Football Athletes During Childhood And Late Adolescence

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There are currently few studies on saccadic eye movements in children and adolescents, especially those with mild traumatic brain injury (mTBI) suffered in a sports related environment. The antisaccade (AS) task in particular may be able to objectively assess for mTBI or the effects of subconcussive loading by comparing baseline to follow-up performance. Since the AS task specifically engages areas such as the prefrontal cortex, quantifying performance may further help to understand which brain regions may be injured during a mTBI or affected due to subconcussive loading. Age-based differences in oculomotor system response resulting from differences in brain development are also of importance when considering the use of an assessment modality across ages.
Participants were males recruited from a high school football team (n = 94; age: 13-18 YRS) and from a Pop Warner football team (n = 34; age: 5-13 YRS). Saccadic eye movement data were collected at baseline and follow-up time points using EyeLink 1000 system (SR Research, Canada). This study is on going and currently analysis of data for the performance within each group on the AS task is still in progress. However, AS metrics analyzed to date include number of wrong-way trials, and preliminary results were based on small sample size of valid data. Baseline data shows that ages 5-13 YRS (Pop Warner participants) completed more wrong-way trials without correction. The trend in this preliminary data shows the younger age group made a greater number of wrong way trials. A greater sample size is needed to assess for statistical difference. Additionally, while many wrong-way saccades were initiated, participants performed corrective saccades in opposition to the target in most cases.

This trend may suggest that the younger group was less likely to inhibit the reflexive prosaccade in accordance with development of the higher-order cortices in the PFC. Alternatively, the trend could be due to a misunderstanding of the antisaccade task by the younger cohort. Overall, this trend of increasing number of correct trials with age matches the findings of previous studies. With further analysis, this research will further our understanding of oculomotor performance in the AS task in relation to development of the brain, especially areas such as the PFC, across different age groups

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Poster Number: 16

The Neurons In Central Neural System Activate And Inhibit Egg-Laying Behavior

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To increase reproductive success, organism evolve many methods. For insect, one important method is to increase the egg rate. Moreover, to provide good environment and increase the survive of offspring, insect also can control the due time even the egg is ready to lay out until they meet good environment. Using drosophila, we find that some good condition can activate egg-laying whereas some bad condition can inhibit egg-laying. We identify a group neurons (bad condition neurons) which are involved in egg-laying controlling by inhibit egg-laying. Those neurons can temporarily control egg-laying behavior. After those neurons lose function, flies can normally lay eggs even in bad condition. Another group neurons(good condition neurons) can process good information and activate egg-laying. Good condition neurons can inhibit bad condition neurons to compete with bad condition.

Poster Number: 17

Cell Type-Specific Contributions Of Ube3a Loss To Epilepsy In Angelman Syndrome

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Epilepsy is caused by both precipitating factors and genetic predisposition. Deletion or mutation of the maternally inherited UBE3A allele, which encodes the ubiquitin protein ligase E3A (UBE3A), results in EEG abnormalities and severe intractable epilepsy in individuals with Angelman syndrome (AS). Deletion or mutation of the orthologous maternal Ube3a allele in mice is also sufficient to enhance seizure susceptibility. However, in both mouse and man, cellular and circuit-level mechanisms of epileptogenesis following UBE3A loss remain to be elucidated. Our study of a mouse model of epilepsy reveal that maternal loss of Ube3a reduces flurothyl-induced seizure threshold in naïve mice. To gain insight into how excitatory or inhibitory neuron contributes to the development of circuit hyperexcitability in AS, I tested flurothyl sensitivity in naïve mice with selective loss of maternal Ube3a from either glutamatergic (Ube3aFLOX/p+::NEX-Cre) or GABAergic (Ube3aFLOX/p+::Gad2-Cre) neurons. Ube3aFLOX/p+::Gad2-Cre mice exhibited an approximately 30% reduction in seizure threshold. In contrast, seizure thresholds were similar between control and Ube3aFLOX/p+::NEX-Cre mice, suggesting GABAergic Ube3a loss contributes to pro-convulsive pathologies of AS. Moreover, by spatio-temporally profiling the post-ictal expression of immediate-early genes, I will trace the specific neural circuits that are most vulnerable to UBE3A loss. Combined, this study will elucidate cellular and possible circuit mechanisms of retractable epilepsy in AS and reveal therapeutic targets.

Poster Number: 18

Cholinergic Regulation Of The Hippocampal Output To Entorhinal Cortex

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The hippocampus is the major brain structure that plays an important role in memory; it receives sensory inputs from the cortex during memory encoding while transferring the temporary hippocampal information back to the cortex during memory consolidation. It has been shown that acetylcholine (ACh) stimulates memory encoding while inhibiting the memory consolidation process, but the underlying mechanism is yet unclear. The CA1 pyramidal neurons in the hippocampus are the final output of the hippocampus and receive feedback inhibition from the GABAergic interneurons oriens lacunosum-moleculare (OLM) cells which express high levels of ACh receptors (AChRs). We examined if ACh modulates OLM interneurons to increase negative feedback in the hippocampus to decrease the hippocampal output to the entorhinal cortex (EC). The modulation is important for proper memory encoding given that deep layer EC neurons projects to the superficial layer EC neurons allowing re-entrance of the processed information back to the hippocampus, which will interfere with new coming information while memory encoding is active
In this study, we used electrophysiological, optogenetic, pharmacological, and behavioral approaches to study how ACh modulates OLM interneurons to regulate the hippocampal output to the EC. Optogenetic stimulation of cholinergic neurons depolarized OLM interneurons, causing an increase in spiking activity of the neurons. The increase in firing of OLM interneurons caused an increase in inhibitory GABA inputs to CA1 pyramidal neurons, suppressing the firing activity of the neurons. Consistently, photostimulation

of either cholinergic or OLM neurons caused a decrease in CA1-evoked currents in the layer V EC (ECV). We demonstrate here that ACh, via acting at OLM interneurons, increases negative feedback in the hippocampus and decreases hippocampal output to the EC. The ACh regulation of OLM interneurons is important for memory encoding of hippocampus-dependent learning, which is impaired when OLM interneurons are ablated via the expression of diphtheria toxin A.

Poster Number: 19

Hypothalamic Involvement In Nicotine Self-Administration In Rats

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The hypothalamus is a brain region that has typically been overlooked regarding its potential contributions to processes of drug addiction. Classic literature has shown hypothalamic involvement in consummatory behavior. Drug addiction shares with feeding regulation key behavioral aspects of the appetitive urge. In addition to its primary role as a regulator of metabolic and autonomic function, the hypothalamus is a limbic structure composed of several distinct nuclei that both project to and receive input from several regions of the brain, including areas involved in memory, attention, emotion, and reinforcement learning. The functional output of these projections is responsive to, and regulated by, dopaminergic, serotonergic, noradrenergic, and cholinergic activity. We are investigating the contributions of hypothalamic nuclei to nicotine addiction in a rat model of nicotine self-administration. The first series of experiments targeted D1 dopamine receptors located in the supramammillary nucleus (SuM) of the hypothalamus; a region that has been implicated in the process of positive reinforcement. Young adult female Sprague-Dawley rats were fitted with jugular catheters and given access to self-administer nicotine (0.03 mg/kg) on an FR1 schedule of reinforcement. Each self-administration session lasted 45 min. Bilateral infusion cannulae were implanted in the SuM to allow local infusion of the D1 receptor antagonist SCH23390. Infusions of SCH23390 occurred 5 min prior to the start of each session. Doses of SCH23390 (1, 2, and 4 µg/side) were infused in a repeated measures, counterbalanced design two times for each rat. Bilateral infusions of SCH23390 into the SuM caused significant reductions in the number of nicotine infusions per session at the 2 ($p<0.05$) and 4 ($p<0.01$) µg/side doses when compared to infusions of the aCSF vehicle. These results demonstrate that hypothalamic D1 dopaminergic innervation plays an important role in the process of nicotine self-administration. Hypothalamic mechanisms may be key components of the neural circuitry underlying addiction. Future experiments will examine the contributions of other transmitters innervating hypothalamic nuclei to nicotine addiction including the involvement of serotonergic and cholinergic receptor mechanisms to this process.

Poster Number: 20

Sustained Trkb Activation Is Required For Maintenance Of Ltp

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Temporal Lobe Epilepsy (TLE) is a devastating neurological disease for which no disease modifying therapy exists that is characterized by spontaneous recurrent seizures. A survey of patients with TLE revealed that 89% had suffered a prolonged seizure during childhood and then experienced a latent seizure free period before the commencement of spontaneous recurrent seizures (French et al., 1993). We believe this latent period presents a therapeutic window during which treatment could reverse the pathology and prevent development of the disease. One hypothesized mechanism of epileptogenesis is the long-term potentiation (LTP) of excitatory synapses between excitatory neurons that occurs during seizure activity. LTP has an electrophysiologic component (eLTP) and a structural component (sLTP). Both eLTP and sLTP are tightly correlated with one another, and both require the release of Brain Derived Neurotrophic Factor (BDNF) and the activity of small GTPases for their induction and maintenance. Using a reduced preparation of organotypic hippocampal slice culture together with two photon glutamate uncaging and FRET based sensors, we are investigating the cellular mechanisms through which BDNF signaling regulates small GTPases in the maintenance of LTP at an individual synapse. Here, we show that sustained activation of the BDNF receptor TrkB is necessary for the sustained activation of the small GTPase Rac1 and the maintenance of sLTP.

Poster Number: 21

Shank3 Deficiency Impairs Pain Signaling Transduction In Primary Sensory Neurons

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SHANK3 is a postsynaptic scaffold protein in the central nervous system. Accumulating studies implicated the critical role of SHANK3 in autism spectrum disorder (ASD). Here we report that the presynaptic expression of SHANK3 in mouse dorsal root ganglion (DRG) sensory neurons and spinal cord dorsal horn involved into heat pain transduction. Shank3 (Δe4-22) deficient mice displayed impaired inflammatory

and neuropathic pain. Specific loss of SHANK3 in Nav1.8-expressing sensory neurons only impairs heat pain but not mechanical pain. Biochemical study showed that SHANK3 interacts with transient receptor potential subtype V1 (TRPV1) via Proline-rich region and regulates TRPV1 membrane trafficking. Furthermore, TRPV1 signaling, including capsaicin-induced peripheral and central pain, DRG inward currents, and spinal cord synaptic currents are all substantially reduced after Shank3 haploinsufficiency. Finally, knockdown of SHANK3 expression in human DRG neurons also abrogates TRPV1 function. Our findings reveal a peripheral and presynaptic mechanism of SHANK3, which may underlie pain deficits in SHANK3-related autism.

Poster Number: 22

Maternal, But Not Paternal, Exposure To Bisphenol A Is Associated With Increased Anxiety-Like Behavior In Juvenile Mice

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Bisphenol A (BPA) is a man-made compound used in the production of polycarbonate plastics and epoxy resins found in many commonly used items. Recent studies in humans and animal models have demonstrated associations between early life exposure to BPA and adverse neurobehavioral outcomes. Our previous research in mice indicates that gestational exposure to BPA (5 mg/kg diet) significantly alters juvenile social interactions, social preference, and social recognition. The goal of the current study was to extend these findings. We fed naïve females a daily pellet 10 days prior to mating and throughout pregnancy containing vehicle or 20 micrograms of BPA, a dose similar to our previous exposure paradigm. We assessed the behavior of juvenile F1 offspring in a social recognition task and on the elevated plus maze. We also exposed naïve adult males to BPA or vehicle in a similar manner for 40 days before pairing with an unexposed female. The F1 offspring from these pairings were tested in the same way as maternally exposed offspring. Contrary to our previous experiments, we found significant differences between maternally exposed F1 BPA mice and controls in the elevated plus maze test, but not in the social recognition test. F1 offspring exposed via the paternal lineage displayed no significant differences from controls in the behaviors tested. Several differences between the two exposure paradigms likely contributed to these conflicting results, like the difference in diet phytoestrogen levels and the lack of fostering at birth. Future experiments should explore the role of these factors in modulating the effect of BPA on juvenile behaviors.

Poster Number: 23

Variations in Response Gain in Frontal Cortex Linked to Variability in Saccadic Reaction Time

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We revisited a fundamental question in oculomotor neuroscience: how does single-neuron activity in the frontal eye field (FEF) relate to the timing of eye movements? To investigate the neural correlates of choice, reward availability, reaction time (RT), and movement metrics, monkeys performed a RT variant of the one-direction-rewarded (1DR) task. In each trial, the animals maintained fixation at a central spot and made a saccade when an eccentric stimulus appeared at one of 4 possible locations, but crucially, only one location was associated with the primary reinforcer. Behavioral effects were clear: saccades to rewarded locations were precise and consistently short latency, whereas those to unrewarded locations were longer latency and of highly variable metrics. We exploited the large spread in RT and spatially distinct reward conditions in the 1DR task to study how individual FEF neurons contribute to saccade production. This exposed a novel, strong dependency: for most neurons, the maximum firing level either increased or decreased monotonically as a function of RT. This was true for all neuronal classes in FEF regardless of their visuomotor properties. Furthermore, modeling results suggest that the two complementary populations with similar response fields but opposite temporal selectivities serve a distinct purpose, to control, according to their relative gain, whether the ensuing RT is short or long. These findings are significant for two reasons. First, it is thought that saccades are triggered when the firing level in FEF reaches a fixed threshold, but according to our results, this is true only in an average sense; for individual cells, the presaccadic firing rate attained may vary substantially with RT, either positively or negatively. Second, the results pinpoint a fundamental source of variability in RTs — fluctuations in the gain of fast- and slow-preferring complementary populations — and propose a specific mechanism whereby cortical circuits may regulate the timing of motor commands.

Poster Number: 24

A Novel Faah Inhibitor Protects Prefrontal Cortex Neurons From Hiv-1 Tat-Induced Excitotoxicity

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Cannabinoids have been shown to be neuroprotective in a variety of neurodegenerative diseases, including Alzheimer's and Parkinson's disease, but little work has been done to explore their potential benefits in neuro-acquired immune deficiency syndrome (neuroAIDS). The advent of combined antiretroviral therapy (cART) has greatly improved the prognosis of patients infected with human immunodeficiency virus type 1 (HIV-1). Unfortunately a significant proportion of this population still exhibit a range of marked decreases in cognitive abilities, often referred to as HIV-1 associated neurocognitive disorder (HAND). HIV-1 does not infect neurons but produces viral toxins, such as transactivator of transcription (Tat), that have been shown to disrupt cellular calcium equilibrium resulting in excitotoxic conditions. The increase in intracellular calcium can give rise to synaptodendritic injuries and cell death, the former which is highly correlated with loss of cognitive function in HIV-1 patients. The purpose of this study was to characterize the neuroprotective benefits of the endocannabinoids, AEA and 2-AG, in Tat treated mouse primary prefrontal cortex cell culture. We conducted calcium imaging and time-lapse experiments to asses cell calcium content and viability after being exposed to Tat with or without additional endocannabinoids present. Our results support prior literature showing that Tat produced excitotoxic levels of intracellular calcium and decreased neuronal viability. Further the direct application of endocannabinoid ligands reduced intracellular calcium and promoted neuronal survival under Tat conditions. Upregulating AEA levels using the highly selective and potent FAAH inhibitor, PF3845, also blunted Tat-induced intracellular calcium increase, which was not noted with an inhibitor of MAGL activity, MJN110. Lastly, pretreatment with the CB1 antagonist SR141716A abolished the neuroprotective effects of AEA, 2-AG and PF3845 suggesting they all exert their effects through the CB1 signaling pathway. Exploring the interactions between Tat toxicity and endocannabinoid signaling has the potential to identify novel therapeutic interventions to benefit individuals suffering from HAND and other cognitive impairments.

Poster Number: 25

Chemogenetic Inactivation Of The Insular Cortex Increases Interoceptive Sensitivity To Alcohol

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Previous work has demonstrated a role for the nucleus accumbens core (AcbC) in regulating the interoceptive/subjective effects of alcohol. In order to investigate potential novel circuits involved in modulating sensitivity to alcohol, male Long-Evans rats (n=12) received a microinjection of the retrograde tracer Fluorogold (FG) directed at the AcbC. Projections to the AcbC were visualized through immunohistochemistry. Dense FG immunoreactivity (IR) was found in the insular cortex (IC). Next, to begin to examine a possible role for IC projections to AcbC (IC⟶AcbC) in modulating sensitivity to alcohol, we examined neuronal response to alcohol (1 g/kg, IG). Examination of co-localized FG and c-Fos IR revealed an alcohol-induced increase in c-Fos IR within FG positive cells, demonstrating an IC⟶AcbC specific response to alcohol. Next, to validate the use of a chemogenetic approach by which to inactivate the IC, rats were microinjected with inhibitory Designer Receptors Exclusively Activated by Designer Drugs (DREADDs; hM4D(Gi)) in the IC. DREADD expression and functional validation were confirmed with IR and electrophysiological recordings. To determine the functional role of the IC in modulating sensitivity to alcohol, male Long-Evans rats (n=6-9) were microinjected with the Gi DREADDs in the IC and trained to discriminate alcohol (1 g/kg, IG) vs. water using standard drug discrimination techniques. Rats received pretreatment with 1 mg/kg clozapine-N-oxide (CNO; IP) to activate the Gi DREADDs, 45 min prior to a cumulative alcohol curve (0.1, 0.3, 1.0, and 1.7 g/kg). Chemogenetic activation of the Gi DREADDs following CNO pretreatment resulted in increased sensitivity to low alcohol doses (0.1 – 0.3 g/kg), demonstrating a role of the IC in regulating the interoceptive effects of alcohol. Further these data demonstrate the feasibility of investigating a striatal-insula circuit in modulating sensitivity to alcohol.

Poster Number: 26

Genetic And Environmental Determinants Of Neonatal Cortical Thickness And Surface Area

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Genetic and environmental influences on brain structure are expected to vary in a complex and highly dynamic way across the human lifespan. While it is established that cortical thickness (CT) and surface area (SA) are largely genetically distinct in adults and that heritability varies across cortical regions, complementary studies in infants have not been performed. This represents a critical knowledge gap, given compelling evidence that neuropsychiatric disorders have their ultimate origin in prenatal and early postnatal development. Using a large sample of twins and singletons, we performed the first comprehensive assessment of the heritability of global and regional SA and CT in infancy. Structural MRI images were obtained from 856 neonates (163 monozygotic twins, 284 dizygotic twins and 409 singletons). Cortical surfaces were reconstructed and parcellated into 90 regions of interest. Cortical thickness was defined as the average value of the minimum distance from the inner to the outer surfaces and the minimum distance from the outer to the inner surfaces.

Surface area was computed based on a geometrically central cortical surface. Linear mixed effect models were used to determine the proportion of total phenotypic variance explained by additive genetic factors as well as proportions due to common and unique environmental factors (ACE model). Heritability was higher for total surface area (0.71) compared to average cortical thickness (0.38). Regional heritability estimates ranged from 0 to 0.52 for cortical thickness and 0.07 – 0.73 for surface area. While less prominent, shared environmental effects ranged from 0.06 to 0.38 for cortical thickness and from 0.03 to 0.48 for surface area. Results indicate that additive genetic factors contribute to both global and regional CT and SA measures in neonates. These findings are consistent with heritability studies performed at later ages and confirm that CT and SA measures are genetically distinct. In addition, our results suggest that unique genetic factors distinguishing surface area and cortical thickness are likely established prior to infancy.

Poster Number: 27

A Gut-Brain Neuroepithelial Circuit

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Enteroendocrine cells, the sensory cells of the gastrointestinal tract, have been studied so far from an endocrine perspective. The reason for this is that enteroendocrine cells were thought to communicate with nerves only indirectly, through hormones such as Peptide YY (PYY), hence, the endocrine portion in their name. However, they have striking features of epithelial cell transducers: they are electrically excitable, fire action potentials, possess voltage-gated channels, express synaptic proteins; and recently, we reported that nerves innervate them. The source of these connections remains to be documented.

Here we use a modified rabies virus to define which peripheral nerves connect with enteroendocrine cells. This rabies virus, RabΔG-EnvA-GFP is unique in two ways: 1) it lacks the rabG protein required to jump a synapse, and 2) it contains the EnvA protein, which means this virus will only infect cells with the Tva surface protein. The virus was delivered by enema into the lumen of the colon of a PyyCRE_RabG::Tva mouse, in which the rabG glycoprotein and Tva surface protein is expressed in PYY enteroendocrine cells. In this way, the RabΔG-EnvA-GFP virus serves to specifically reveal a monosynaptic circuit in enterendocrine cells.

Our results show that PYY enteroendocrine cells are infected by the rabies virus. And, if those cells express the complementary rabG glycoprotein, then neurons of nodose ganglia are labeled. These data revealed that colonic enteroendocrine cells form a monosynaptic neural circuit with vagal nerve fibers. This neuroepithelial circuit offers a path for sensory information to be transmitted directly to the brain and for the brain to respond to sensory stimuli from the gut lumen.

Poster Number: 28

Cb1r Antagonist Treatment Reversed Acute-Binge Ethanol Induced Increase In Hmgb1 And Rage Expression In The Prefrontal Cortex Of Rat

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Background: Recently neuroimmune gene RAGE (Receptor for Advanced GlycationEnd products)) and Toll like receptor4 (TLR4) as well as their endogenous agonist, HMGB1 (high-mobility group box 1) has been implicated in alcohol addiction. Studies from different laboratories have found that chronic ethanol treatment persistently upregulated RAGE, TLR4, HMGB1, and that alcohol dependence is associated with increased expression of these neuroimmune genes. However, in spite of increased incidence of alcohol and cannabinoid exposure during adolescent and adulthood, the effects of combined ethanol and cannabinoid treatment on these neuroimmune genes has not been studied .In the current study using acute binge-like alcohol drinking model, we have investigated a) the effect of ethanol and CB1 cannabinoid receptor (CB1R) agonist (ACEA) on the expression of RAGE and HMGB1 in adult the prefrontal cortex and b) the role of CB1R antagonist SR141716A (SR1) on ethanol and ACEA-induced effects on the expression of RAGE and HMGB1.
Method: Adult male Wistar rats (body wt. 350 -380gms) were treated with vehicle or binge level ethanol (5 g/kg, ig) or CB1R agonist ACEA (3 mg/kg, i.p.) alone or in combination with CB1R antagonist SR141716 (SR1; 3 mg/kg, ip). For antagonist treatment, SR1 was administered 30 min prior to ethanol or ACEA treatment and animals were sacrificed 3hr.following the last treatment. The effects of the ethanol and cannabinoid treatment on HMGB1 AND RAGE expression in the prefrontal cortex was assessed immunohistochemically.
Results & Conclusion: We found that acute binge-level treatment with ethanol or CB1R agonist ACEA alone and in combination significantly increased HMGB1 and RAGE expression in the prefrontal cortex region of the brain. Interestingly, co-treatment with CB1R antagonists (SR141716) significantly reversed ethanol or ACEA induced increase in HMGB1 and RAGE expression. Together, these findings clearly suggest that CB1R plays a key regulatory role on EtOH -mediated effects on HMGB1 and RAGE expression in the prefrontal cortex region of the brain. Thus targeting CB1R for regulation of Ethanol-mediated effects on these neuroimmune factors may have therapeutic potential in alcohol pathobiology in brain.

Poster Number: 29

Human Umbilical Cord Tissue-Derived Cells (Hutc) Promote Synapse Formation And Neurite Outgrowth Via Thrombospondin Family Proteins

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Age-related macular degeneration (AMD) is the leading cause of vision loss in people over the age of 50. Subretinal administration of hUTC to a rodent model of retinal degeneration preserved photoreceptors and visual function. This effect of hUTC is mediated through paracrine effects rather than by trans-differentiation of hUTC into different cell types. However, the therapeutic mechanism of hUTC has not been defined. We hypothesize that hUTC provide therapeutic factors that promote neuronal survival, neurite outgrowth and synaptogenesis. To test this hypothesis we utilized a purified rat retinal ganglion cell (RGC) culture system. In this in vitro system we studied the effects of hUTC-conditioned media on RGC survival, neurite outgrowth and synapse formation. We found that hUTC promote neuronal survival and enhance neurite outgrowth. Interestingly, hUTC strongly induce excitatory synaptogenesis between RGCs as determined by immunolabeling of synaptic markers and electrophysiological recordings. The synaptogenic factors are larger than 100kDa and can be blocked by the antiepileptic drug Gabapentin. Gabapentin is a known blocker of synaptogenic Thrombospondin (TSP) family proteins. We found that hUTC secrete TSPs (TSP1, 2 and 4) and silencing TSP expression in hUTCs eliminated the synaptogenic effects of these cells as well as their ability to promote neurite outgrowth. Our results show that through paracrine effects hUTC enhance neuronal survival, neurite outgrowth and promote development of functional synapses. We identified the TSPs as the major synaptogenic factor secreted by hUTC. hUTC-secreted TSPs also supported neurite outgrowth. Our findings demonstrate that hUTC affects multiple aspects of retinal cell health and connectivity and each of these paracrine effects may individually contribute to the therapeutic function of these cells. These results also reveal that TSP-mediated synaptogenesis might provide a potential therapeutic target in retinal degenerative diseases.

Poster Number: 30

Selective Behavioral Effects Of Trkb-Plcg1 Uncoupling In Epileptogenesis Versus Fear Conditoning

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Epilepsy is a common and debilitating neurological disorder for which no disease-modifying treatments exist. Of the various forms of epilepsy, Temporal Lobe Epilepsy (TLE) is of particular interest because (1) TLE is the most common form of epilepsy; (2) TLE is frequently associated with psychiatric co-morbidities such as anxiety disorder; and (3) TLE is frequently refractory to medical therapy. Studies have linked the development of TLE to a previous episode of prolonged seizure activity (status epilepticus, aka SE). We previously demonstrated a critical role for TrkB-PLCg1 signaling in SE-induced TLE, and designed a peptide that inhibits TrkB-mediated activation of PLCg. This peptide prevents epilepsy when administered after SE, raising the possibility of the therapeutic efficacy of uncoupling TrkB-PLCg1 in preventing epileptogenesis. Of concern, studies using genetically modified TrkB-PLCg1 mice in a fear-conditioning paradigm raise the possibility of anterograde amnesia as an unwanted side effect. We demonstrate that using our peptide at doses higher than that needed to prevent epileptogenesis produces no impairments in fear learning.

Poster Number: 31

Electrochemical Measurements Of Real-Time Opioid Peptide Fluctuations Within The Striatum And Midbrain Of The Rat: A Neuromodulatory Role For Cocaine-Induced Methionine Enkephalin Dynamics

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Neuropeptides regulate a broad spectrum of biological functions, from pain and analgesia to hedonic and motivational behaviors associated with reward and addiction. Opioid neuropeptides modulate the mesolimbic and nigrostriatal dopamine (DA) circuits, which are affected by common drugs of abuse, such as cocaine. Cocaine administration elicits striatal DA release and locomotor sensitization, and several key aspects of cocaine abuse require intact opioid signaling. However, the precise mechanisms that underlie opioid modulation of DA systems remain ambiguous. Although several methods exist for monitoring DA fluctuations, few tools are available for selectively monitoring dynamic fluctuations of endogenous opioid neuropeptides. This work addresses this issue by employing a completely novel electrochemical approach to monitor sub-second fluctuations in methionine-enkephalin (M-ENK) in rat brain tissue. By combining multiple scan rate voltammetry with constant-potential amperometry in every voltammetric sweep, we have measured putative M-ENK fluctuations evoked by cocaine administration in the striatum of an L-DOPA treated 6-OHDA lesioned rat. Given opioid signaling is implicated in the mechanisms that can accompany robust increases in extracellular DA, such as those elicited by L-DOPA or cocaine, our study investigates cocaine-induced effects on in-situ M-ENK transients. Importantly, our electrochemical approach enables selective

detection of chemical species within each scan, including striatal DA release. As such, on-going work is evaluating simultaneous M-ENK and DA dynamics in the striatum, as well as the midbrain (ventral tegmental area / substantia nigra), following acute and repeated administration of cocaine. These measurements of opioid peptide dynamics will complement DA related research on chemical mechanisms that underlie cocaine addiction.

Poster Number: 32

Alterations In Neuronal Function In Central Nucleus Of Amygdala In Bk Beta-1 Or 4 Knockout Mice Following Chronic Intermittent Ethanol Exposure

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Large conductance Ca++ activated potassium (BK) channels associate with specific auxiliary subunits (beta1-4) and are expressed in central neurons. These BK channels regulate neuronal excitability and synaptic transmission. Neurons in the central nucleus of the amygdala (CeA) are involved in alcohol addiction and BK channels are thought to be potential targets for ethanol action as presynaptic BK channels in the CeA mediate alcohol-induced GABA release. The accessory beta1 and 4 subunits of BK channels are essential for modulating BK channel function and sensitivity to actions of ethanol; thus alterations in these subunits may influence the function of CeA neurons after both acute and chronic intermittent ethanol (CIE) exposure. We have examined the membrane properties and inhibitory synaptic neurotransmission in CeA neurons from BK beta1 or beat-4 KO and WT male mice following CIE exposure via a vapor inhalation chamber. Under whole-cell current clamp conditions, several distinct types of firing patterns were recorded in neurons from these mice. No statistical differences in the resting membrane potentials between CeA neurons from WT and KO mice after CIE were observed. In CIE-mice, the mean input resistance of neurons from beta4 KO mice was significantly higher than in neurons from WT mice. The threshold of action potentials was not altered in CeA neurons lacking either beta1 or beta4 in CIE-mice. In response to depolarizing current injections, beta1 KO CeA neurons fire more spikes than WT neurons in CIE-mice but beta4 KO CeA neurons appear to fire fewer spikes than WT neurons after CIE. Upon a strong depolarization, a depolarization block was also observed in some beta4 KO CeA neurons from CIE-mice. Under whole-cell voltage clamp recording, both eIPSCs and sIPSCs were recorded in CeA neurons from beta1 KO and WT CIE-mice. There was a significant increase in input/output curves of eIPSCs evoked by electrical stimulation in beta1 KO CeA neurons of CIE-mice compared to WT CIE-mice. However, no significant differences in the paired pulse ratio of eIPSCs or the mean frequency of sIPSCs of CeA neurons between beta1 KO or WT CIE-mice were detected. Our current results suggest that different BK channel accessory subunits in CeA neurons could regulate neuronal function following long-term intermittent exposure to ethanol, thus providing novel targets for pharmacotherapies aimed at reducing excessive ethanol consumption

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Poster Number: 33

The Effect Of Nasal Oxytocin On Dopamine Release

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The oxytocin and dopamine systems are highly intertwined and have been implicated in a number of similar behaviors, particularly social, sexual, and maternal behaviors. However, the basic pharmacological effect of oxytocin administration on the dopamine system is yet to be determined. Additionally, although nasal oxytocin is the primary route of administration in human research, it is rarely used in animal studies, with researchers often relying on other routes for systemic oxytocin administration (e.g., intraperitoneal injection). The present study aims to fill this gap in the literature by examining the pharmacological effect of nasal oxytocin on the dopamine system using fast-scan cyclic voltammetry. Specifically, stimulated dopamine release in the nucleus accumbens was measured before and after nasal oxytocin administration in female rats. Because the peripartum period is rife with neuroendocrine changes, both virgin and postpartum animals were used to examine how pregnancy affects oxytocin-dopamine interactions. Postpartum rats exposed to cocaine during pregnancy were also used, due to gestational cocaine's deleterious effects on both the oxytocin and dopamine systems. Although data collection is still underway, there does not appear to be a clear effect of oxytocin on dopamine release, which is inconsistent with past studies in the field. This may be due to any of the critical characteristics of this project – anesthetized animals, stimulated release, and phasic dopamine – which differ from the studies used to inform the project's design.

Poster Number: 34

Posttranscriptional Regulation Of Embryonic Neurogenesis By The Exon Junction Complex

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The mammalian neocortex is the brain structure responsible for higher cognitive function. The neocortex is built via the generation of neurons by neural stem cells in a process termed neurogenesis. Alterations in stem cell function during neurogenesis can lead to atypical neocortex

formation, impairments to cortical functions, and microcephaly, in which brain size is significantly reduced. Here we study the role of the exon junction complex (EJC), a protein complex required for several RNA processing events, in mouse embryonic neurogenesis. The EJC core consists of three proteins, Magoh, Rbm8a, and Eif4a3. The EJC regulates RNA splicing, subcellular localization, translation and nonsense mediated RNA decay (NMD). Mutations in EJC core and peripheral components are strongly associated with human neurodevelopmental pathologies. Yet the underlying mechanism by which EJC levels impact brain development and disease remain unknown. We generated conditional mouse models of all three EJC core components. We find haploinsufficiency of individual EJC core components phenocopy each other and cause microcephaly due to reduced neuron stem cells, ectopic production of neurons, and extensive apoptosis. We use mouse genetics, transcriptome and proteome analyses to demonstrate haploinsufficiency for any of the three core EJC components disrupts pathways critical for translation regulation and cell viability. In addition, we find that p53 is activated in EJC haploinsufficient NSCs and p53 depletion significantly rescues EJC-mediated microcephaly. Altogether our study demonstrates translation and p53 activation are major nodes of EJC-dependent cortical development regulation, suggesting potential mechanisms for the etiology of EJC associated neurodevelopmental diseases

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Poster Number: 35

Prenatal Choline Supplementation Exhibits Sexually Dimorphic Anti-Inflammatory Effects In A Mouse Model Of Prenatal Diesel Exposure

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Numerous studies have shown that air pollution causes widespread inflammatory processes in body and brain and is linked to neurocognitive difficulties, increased anxiety and depression, and increased prevalence of neurodegenerative disorders. When exposure to air pollution occurs early in development, children show decreased working memory ability (Sunyer et al., 2015). As well, prenatal exposure to diesel particulate matter increases inflammatory cytokine expression within several brain regions of embryonic day 18 males and leads to long-term negative outcomes for the offspring (Bolton et al., 2012; 2014). In contrast, dietary choline supplementation has been shown to decrease inflammation in adult rats and humans (Rivera et al., 1998; Mehta et al., 2010) and when administered as a supplement to pregnant rats, choline also increases working memory and decreases age-related cognitive decline in the offspring (Meck et al., 2008). The current study sought to determine if dietary choline supplementation protects against the deleterious effects of air pollution on the developing brain. Time-mated C57/Bl6 mice were given a high-choline (SUP) or control (CON) diet, and a series of diesel particulate (DEP) or vehicle (VEH) exposures throughout pregnancy. Mice were sacrificed and tissues were collected on embryonic day 18. The number and activation state of microglia, identified by Iba1+ immunohistochemical staining, in several brain regions were examined to determine the impact of choline and/or diesel on microglial development. Additionally, qPCR was performed to assess expression of genes associated with inflammation. We found that male CON/DEP mice had more activated microglia in the dentate gyrus than the CON/VEH and SUP/VEH groups. Prenatal choline supplementation to the DEP group completely prevented this increase in microglia number and change in morphology. Remarkably, females do not show these effects in response to diesel or to choline. The effects of choline appear to be regionally specific between hippocampus, hypothalamus, and amygdala. These findings suggest that prenatal choline supplementation throughout pregnancy may protect the fetal brain against the neuroinflammation associated with diesel air pollution exposure, and the effects of both diesel and prenatal choline supplementation are sexually dimorphic. Further work is underway to determine how choline supplementation to the pregnant dam leads to alterations in fetal and maternal responses to prenatal air pollution.

Poster Number: 36

Early Postnatal And Adult Neurogenesis In The Forebrain Differentially Regulate Detection Of Novel Aversive And Appetitive Odors In Mice

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Generation of new neurons in the central nervous system (CNS) is prevalent during embryonic development. Postnatal and adult neurogenesis are limited to distinct regions of the mouse CNS including the hippocampus and the olfactory bulbs (OB). New neurons in the OB arise from neural stem cells in the subependymal zone (SEZ) and the rostral migratory stream (RMS) from where they migrate followed by differentiation and integration into existing circuits. Despite substantial progress over the last two decades in identification of cellular and molecular mechanisms that regulate adult OB neurogenesis, the physiological significance of continuous integration of newborn neurons in the adult OB remains unclear. This lag is largely due to the absence of tools that allow for assaying the role of mosaic neuronal populations in the OB without invasive manipulation. Here we report implementation of Designer Receptors Exclusively Activated by Designer Drugs (DREADDs) to inhibit newborn neurons in early postnatal and young adult OBs. In young adults, DREADDs-mediated

inhibition in a fraction of newly generated interneurons profoundly disrupted detection of novel appetitive odors, whereas detection of a novel aversive odor remained unperturbed. Alternatively, detection of a novel aversive odor was significantly abolished upon inhibition of a cohort of neurons born during the first week of postnatal life in mice. Our findings implicate differential age-dependent roles for newborn neurons in the detection of novel appetitive and aversive odors.

Poster Number: 37

Pathway-Specific Striatal Substrates For Habit

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The striatum receives inputs from many areas of the brain and conveys action-related information to the basal ganglia via the direct and indirect pathways. It remains unclear how the propagation of action potentials through direct and indirect pathway striatal projection neurons might change with experience. It has long been thought that an imbalance between these pathways could alter behavior, but relativistic measurements have proven elusive. Using two-photon population calcium imaging and transgenic reporter mice to measure evoked action potential firing in direct and indirect pathway striatal projection neurons simultaneously, we examine how habit alters dorsolateral striatal activity. The dorsolateral striatum (DLS) is required for habit formation and altered neural activity has been observed in this brain region in vivo as habits develop. Mice were trained in an operant lever press task to express or suppress habitual responding. Habitual behavior was associated with increased gain in both DLS output pathways as well as a tendency for direct pathway SPNs to fire before indirect pathway SPNs. The ability to suppress a learned habit was solely predicted by weakened direct pathway output. All effects were broadly distributed across imaged neurons. Together, these findings indicate that the striatum imposes broad, pathway-specific modulations of incoming activity to promote and suppress habitual execution of learned motor behaviors.

Poster Number: 38

Effects Of Vitamin D Receptor-Acting Compounds On Larval Motility In Zebrafish

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Vitamin D is important for a wide variety developmental processes in addition to ion homeostasis and bone growth. Critical among these roles is proper neurodevelopment. Toxicants that disrupt vitamin D signaling during brain development can produce neurobehavioral impairment. In this study we used the zebrafish model to provide an initial characterization of chemicals that act on the vitamin D receptor (VDR) during neurodevelopment and their effects on neurobehavioral function. Using a larval motility assay, the swimming activity of five-day old larvae was assessed by computer video analysis (DanioVision, Noldus, Inc.) in 96-well plates under alternating conditions of dark and light over a 50-min session in 10-min periods. Activity was recorded as mm/min. Testing took place on day five post-fertilization after treatment via immersion to vitamin D (0.5 and 2 nM) and select VDR antagonists PS121912 (2.5 and 5 µM), KT112213 (2, 5 and 10 µM), K-dicyanoaurate, cadmium HCl, cadmium dinitrate (5 and 10 µM each), and 0.1% DMSO control. There were 13-48 subjects per treatment condition and 87 controls. Increased lethality was seen with 5 and 10 µM KT112213 as well as 5 and 10 µM K-dicyanoaurate. Increased dysmorphogenesis was seen with 1 and 2 nM vitamin D, 2 and 5 µM KT112213, 2 and 5 µM K-dicyanoaurate and 10 µM cadmium HCl. Significant larval locomotor hyperactivity was seen with 2 nM vitamin D, 5 µM PS121912 (in the dark phase), 10 µM cadmium HCl, and 5µM cadmium dinitrate (in the dark phases). Locomotor hyperactivity was not consistently related to dysmorphogenesis, and even with the vitamin D- and cadmium HCl-exposed larvae, it appears unlikely that malformations would themselves cause hyperactivity. In contrast, significant larval hypoactivity was seen with 5 µM KT112213, which may be related to increased dysmorphogenesis. Further research will focus on persistent neurobehavioral effects as well as molecular and cellular mechanisms.
Supported by the US-EPA Star grant 83554101

Poster Number: 39

The Ox1r Is The Primary Orexin Receptor Subtype Within The Vta And Central Amygdala That Selectively Modulates Binge-Like Ethanol Drinking

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Although centrally synthesized exclusively within the hypothalamus, orexin (OX) neurons project to various structures throughout the brain and act on two receptors, OX1R and OX2R, to modulate a host of physiological functions including reward and stress- both of which are important factors that contribute to the motivation to consume alcohol. We and others have recently reported that binge-like ethanol drinking leads to changes hypothalamic OX levels (Olney et al., 2015) and that peripherally administered OXR antagonists curtail binge-like ethanol drinking (Anderson et al., 2014); however, these studies were unable to address the specific OX pathways

that modulate this behavior. Thus, in an effort to better characterize this circuitry, the present study was designed to identify the source of OX signaling and elucidate the participation of each OXR within specific brain regions involved in reward- and stress-processing. The "drinking-in-the-dark" paradigm was used to assess changes in the OX system in distinct hypothalamic subregions following binge-like ethanol drinking. We also assessed binge-like drinking of ethanol or sucrose following bilateral infusion of either the selective OX1R antagonist, SB-334867, or the selective OX2R antagonist, TCS-OX2-29, directly into the VTA or CeA of C57BL/6J mice. Results suggest that binge-like ethanol drinking leads to increased OX signaling- particularly within the lateral hypothalamus. We also observed that inhibition of the OX1R, but not OX2R, selectively reduced binge-like ethanol drinking in both the VTA and CeA. Moreover, subsequent investigations revealed that such manipulations did not alter anxiety-like behavior as measured via an open-field locomotor test or the elevated-zero maze. Interestingly, contrary to previous studies that delivered similar compounds systemically, we found no evidence that signaling onto the OX1R in these regions modulates binge-like sucrose consumption; furthermore, we also observed that the OX2R in the VTA and CeA likely does not significantly participate in binge-like ethanol drinking. Together, these data demonstrate that OX- released from the lateral hypothalamus- acts on OX1Rs in the VTA and CeA to modulate binge-like ethanol drinking behavior and that separate OX circuitry governs responding to distinct reinforcers. What is more, the fact that these manipulations did not impact anxiety-like behavior suggests that the OX system likely modulates binge-like ethanol drinking via reward-related circuitry rather than stress-related circuitry. (Supported by NIH grants AA022048, AA013573, & AA015148).

Poster Number: 40

The Effects Of Cholinergic Manipulations On Adult Neurogenesis

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Adult neurogenesis springs from a pool of readily available slowly-renewing neural stem cells (NSCs). NSCs in the hippocampus express nestin, an intermediate filament which can be used to identify them. The cells are quiescent due to tonic GABAergic spillover-stimulation from nearby parvalbumin interneurons. When activated, they divide into intermediate progenitors (IP) or astrocytes. IPs transiently express Tbr2 and MCM2 and differentiate to form new neurons, that in turn express double-cortin (DCX). The process of maturation of a new neuron can take up to 8 weeks and is thought to replicate neurogenesis in a developing pup.

We hypothesized that cholinergic innervation, important during neurogenesis, also affects adult neurogenesis and specifically that the α7nAChR is neuroprotective of pools of neural stem cells in the subgranular zone of the dentate gyrus available for adult neurogenesis. To investigate this we crossed Nestin-creERT2 mice to a ROSA-COP4tdTomato mice, causing the nestin+ NSCs in the mouse to fluorescence red subsequent to injection of Tamoxifen. Prior work has suggested that maternal exposure to a high choline diet increases neurogenesis, whereas nicotine can reduce BrdU positive new neurons and loss of the nicotinic β2 subunit also results in lower levels of neurogenesis as measured by BrdU. To determine whether chronic exposure to nicotine and/or choline would have a bearing on the NSC population, we treated both developing pups and adults and then quantified the nestin+ NSCs and IPs. To look specifically at the effects of loss of α7nAChRs, we quantified the density of nestin+ NSCs in nestin-creERT2 X ROSA-COP4TdTomato X α7nAChR knock-out mouse.
Our research indicates that in juveniles neither nicotine nor choline has a significant quantitative effect on the pool of nestin+ NSCs. However, in adults chronic nicotine reduces density of nestin+ NSCs. Furthermore, giving mice choline during gestation/nursing protects against effects of nicotine in the adult. In the α7nAChR knock-out, both juveniles and adults had significantly fewer nestin+ NSCs. This supports the α7nAChR as neuroprotective of nestin+ NSCs.
The α7nAChR knock-out mice also showed an increased incidence of a subset of cells that are nestin+ ectopic mature granule cells (co-express Prox1 and NeuN, markers of mature granule cells, and morphologically appear to be mature neurons, yet are located deep in the granule layer).

Poster Number: 41

Determining the Sources That Contribute to Extracellular Hydrogen Peroxide Dynamics in the Striatum

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A variety of cellular processes occurring in the central nervous system (CNS) are involved in the generation and accumulation of extracellular Hydrogen Peroxide (H_2O_2). In dopaminergic neurons, generation of H_2O_2 through disturbances in mitochondrial activity as well as the metabolically driven synthesis and breakdown of dopamine (DA) have all been implicated in oxidative stress (Avshalumov et al., 2007; Bao et al., 2009; Patel & Rice, 2012). The goal of this project is to test possible sources of extracellular H_2O_2 by manipulating various pathways through utilization of pharmacological agents. The drugs used for this study will alter processes either directly or indirectly involved in H_2O_2 fluctuations: (1) the DA synthesis and breakdown pathways and (2) the mitochondrial electron transport chain. Through utilization of an m-phenylenediamine (mPD) membrane applied to one carbon

fiber microelectrode (CFM) in a dual microelectrode device, we can directly distinguish H2O2 from other interfering analytes that oxidize at similar potentials. On the uncoated CFM, we can determine what effects these various drugs may have on DA dynamics. During this study, we utilized local microinfusions of pharmacological agents (mercaptosuccinic acid, 735 pg and Triazole, 345 pg) into the tissue next to the recording site, as well as systemic administrations (levodopa, 100 mg/kg and pargyline, 75 mg/kg) that target the entire body of the rat while recording in the striatum. Results from this study will provide information on what cellular processes are responsible for the generation and accumulation of extracellular H2O2 and which of these processes contribute the most, if at all, to oxidative stress in the dorsal striatum.

Poster Number: 42

Rapid, Diffuse And Transient Activation Of Calcium/Calmodulin Dependent Kinase 1 Is Required For The Transient Phase Of Long Term Potentiation

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Structural long-term potentiation (sLTP) depends on calcium entry through N-methyl-D-aspartate (NMDA) receptors. Upon entry into the cell, calcium binds and activates calcium/calmodulin dependent kinases (CaMKs). CaMK activity initiates several signaling cascades essential for sLTP, and it is likely that different CaMKs exhibit different spatial and temporal patterns of activation in response to sLTP induction. Although the spatiotemporal dynamics of the activity of one CaMK - calcium/calmodulin dependent protein kinase 2 (CaMKII) - have been studied, the activation profiles of related CaMKs such as calcium/calmodulin dependent protein kinase 1 (CaMKI) are unknown. In this study, we develop a Försters resonance energy transfer (FRET) based sensor to measure the spatiotemporal patterns of one of the CaMKs, calcium/calmodulin dependent protein kinase 1 (CaMKI). Using the sensor, we demonstrate that CaMKI activation during sLTP induction in a single dendritic spine is rapid and transient, and the activity spreads from the induced spine to the nearby dendritic segment. We demonstrate that NMDA receptor activation is necessary for CaMKI activity. CaMKI phosphorylation by an upstream CaMK, calcium/calmodulin dependent protein kinase kinase (CaMKK) is not necessary for activation, but is required for the spreading of CaMKI activity from the spine to the adjacent dendritic segment. Finally, we show that CaMKI activity and CaMKI phosphorylation are required for the transient increase in spine volume immediately following sLTP. The spatiotemporal pattern of CaMKI is distinct from CaMKII, since CaMKII activation is slower, more sustained and restricted to the spine. In addition, the two CaMKs regulate different phases of sLTP – CaMKI activity is required for rapid spine enlargement immediately following sLTP induction, whereas CaMKII activity is required for the sustained spine enlargement several minutes later. The transient phase of sLTP has not been well characterized, and genetic manipulations of CaMKI may be a useful tool to manipulate and study the functions of the transient phase of LTP.

Poster Number: 43

Voltammetric Detection of Amino Acids for the Selective Identification of Methionine-Enkephalin In Vivo

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Endogenous neuropeptides naturally produced in the central nervous system and in the periphery, such as in the adrenal gland. These molecules bind to μ, δ or κ opioid receptors and are comprised of sequences of amino acids. Opioid neuropeptides are implicated in a variety of functions that are necessary for survival including motivation, nociception, emotion, response to stress and feeding behavior. Moreover, these peptides contribute to the rewarding effects of several drugs of abuse. However, the specific role for opioid peptides in these functions remains ambiguous, largely because they are difficult to detect in vivo. As a result, relatively little is known about when and where opioid peptides are released, or the precise physiological conditions necessary for their release and clearance. This work focuses on assessing the selectivity of a novel voltammetric detection strategy for monitoring methionine-enkephalin and similar neuropeptides in vivo. Our lab has demonstrated the utility of our approach by combining fast-scan cyclic voltammetry (FSCV) with constant potential amperometry to detect the opioid peptide methionine-enkephalin. Since all endogenous peptides are comprised of strings of amino acids, it is important to distinguish the individual electroactive contributions of these amino acids in order to reliably monitor peptides within a complex biological environment such as the brain. Currently, we are systematically characterizing the voltammetric signatures of individual amino acids and chain combinations of these building blocks in vitro. The end result will provide a foundation for the reliable voltammetric identification of individual neuropeptides in vivo and in real time.

Poster Number: 44

Thrombospondin Receptor Alpha-2-Delta-1 Controls Synapse Formation And Maturation In The Developing Visual Cortex

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Astrocytes, the most abundant cell type in the brain, have been identified as key regulators of synaptogenesis. Astrocytes secrete thrombospondin family proteins (TSPs) that strongly induce excitatory synapse formation between cultured retinal ganglion cells (RGCs). Our lab has previously shown that TSP acts through the neuronal calcium channel subunit, alpha-2-delta-1. However, the mechanism by which this interaction results in synapse formation is unknown. We have recently shown that another astrocyte-secreted protein, hevin, specifically regulates thalamocortical synaptic input in the developing visual cortex. This study was the first to show that astrocytes play a role in the establishment of circuit connectivity between brain regions. Taking a similar approach, we have now found that alpha-2-delta-1 is required for the proper establishment of intracortical synapses both in vitro and in vivo. In addition, lack of alpha-2-delta-1 function results in significant deficits in both dendritic outgrowth and spine morphology, revealing the importance of this protein for numerous stages of neuronal development. Finally, we have begun to uncover the cellular mechanism for TSP/alpha-2-delta-1-induced synaptogenesis by assessing downstream signaling of the small Rho GTPase, Rac1.
In summary, our results show that alpha-2-delta-1 is a critical regulator of synaptic connectivity and morphology in the developing visual cortex

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Poster Number: 45

Repeated Intermittent Ethanol Exposure In Adolescent Rats Promotes Enduring Changes In Astrocyte Signaling

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Excessive and repeated alcohol use is highly prevalent in adolescents and young adults and has been associated with an increased risk of developing an alcohol use disorder later in life. The impact of alcohol consumption during this timeframe is critical to understand because it coincides with neuronal maturation, highlighted by the pruning of synaptic connections between neurons and the refinement of circuitry crucial for memory processing, planning, and inhibitory control. We have recently reported that rats exposed to adolescent intermittent ethanol (AIE, 5g/kg i.g. intermittently over 16 days) show long-lasting changes to hippocampal synaptic plasticity and dendritic spines in adulthood that suggest heightened synaptic plasticity and functional immaturity. The timing of these changes corresponds with the dysregulation of a family of astrocyte signaling proteins called thrombospondins (TSPs) and the neuronal synaptogenic receptor α2δ-1 with which they interact. TSPs are involved in synaptogenic processes during development and after injury, and they have been implicated in the development of aberrant synaptogenesis and signaling. Our ongoing work aims to understand the role of astrocyte signaling factors in mediating the long-lasting changes to hippocampal structure and function after AIE.
Our most recent work using Western blot and immunohistochemistry has shown that the astrocyte-secreted matricellular protein hevin (also known as secreted protein acidic and rich in cysteine (SPARC)-like 1) is upregulated in the hippocampus in response to AIE. As with TSPs, this signaling protein is known to be involved in excitatory synapse formation and has been shown to play a pivotal role in the development of appropriate excitatory synaptic connections. This is an exciting finding since hevin has been implicated in the maturation of synapses formed under the influence of TSPs. Work is ongoing to determine whether SPARC, known to antagonize hevin's synaptogenic effect, is also upregulated, thereby preventing the maturation of dendritic spines. These data suggest that astrocyte-related signaling may be contributing to the changes in neuronal circuitry and function previously reported.

Poster Number: 46

Medial Prefrontal Cortex Corticotropin-Releasing Factor Modulates Binge-Like Ethanol Consumption

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Corticotropin-releasing factor (CRF) is a 41-amino acid poly-peptide widely expressed throughout the brain. Multiple lines of evidence support a role of alterations in the CRF system in the transition to alcohol dependence and in relapse to alcohol seeking/consumption during withdrawal. The medial prefrontal cortex (mPFC) plays a critical role in drug and alcohol-dependence. Further, CRF signaling within the mPFC modulates anxiety-like behavior. As neural circuits involved in anxiety-like behavior are well-known to influence drug seeking and intake, mPFC CRF activity is well situated to modulate alcohol related behaviors. Within this work we evaluated the role of mPFC CRF activity in binge-like alcohol consumption in non-dependent animals through use of the drinking in the dark (DID) model. The role of the CRF receptor 1 (CRFR1), which servers as the primary CRF receptor within the mPFC, was specifically evaluated. The mPFC of singly housed C57BL/6J male mice (Jackson Lab, Bar Harbor, Maine) were bilaterally cannulated. Following a week of recovery, mice underwent DID alcohol/sucrose exposure. On Tuesday-Thursday of each week water bottles were removed 3.5 hours into the dark cycle and replaced with sipper tubes containing a 20% ethanol or 3% sucrose solution for a 2 hour period. At the end of this time changes in sipper tube volume were assessed and water bottles replaced. Each Friday this DID period was extended to 4 hours of drinking and ~30 minutes prior to DID session start animals received a microinjection of either a vehicle (95%

saline and 5% DMSO) or CRFR1 antagonist (Antalarmin (Cayman Chemicals, Ann Arbor, MI) 0.5 µg/µl in 95% saline and 5% DMSO) solution using a Latin Square design in which mice experienced each dose of drug in a counterbalanced order (1 DID cycle per dose). Antagonizing mPFC CRFR1 significantly reduced alcohol consumption at the 1 and 4h time point and further significantly reduced blood ethanol concentration at the end of the 4h session. This treatment was also found to reduce sucrose consumption at the 3 and 4h, but not 1h, time point. These results suggest mPFC CRFR1 modulates the consumption of rewarding substance in a time-dependent manner. Further evaluation of mPFC CRF signaling will be of great interest in determining the neural underpinnings of alcohol dependence. Supported by NIH grants AA022048, AA013573, & AA015148.

Poster Number: 47

Novel Animal Models Of Initial Cocaine Sensitivity Using Collaborative Cross Mice

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Substance use disorders (SUDs) are highly prevalent and impose a substantial burden on society. Although these disorders are highly heritable, identifying specific genes and mechanisms in humans has been difficult due to symptomatic heterogeneity and inability to perform baseline assessments in humans after prolonged drug exposure. Animal models of specific aspects of SUDs allow for assessment of underlying mechanisms and genetics. Novelty-induced locomotion has been used to predict initial sensitivity and self-administration of psychostimulants. However, the link between these behaviors has varied across studies and identification of underlying mechanisms has been challenging.
We identified two Collaborative Cross Recombinant Inbred Intercross (CC-RIX) lines that showed significantly high (CC-RIXhigh) and low (CC-RIXlow) locomotor response to novelty. We predicted and confirmed that initial locomotor response to cocaine is also higher in CC-RIXhigh animals compared to CC-RIXlow. We believe these two strains can be utilized as models of high and low predisposition for initial drug sensitivity. We have designed a set of experiments to further characterize these strains for a range of addiction-related behaviors and begin to elucidate the underlying mechanisms for these phenotypic differences. These studies include investigation of stress response, dopaminergic pathways and pharmacokinetics. Additionally, we are using the unique genetic makeup of CC lines to map candidate regions underlying divergent cocaine responses. We believe that utilizing Collaborative Cross mice will enhance our ability to examine the link between initial drug use and progression to addictive-like behaviors due to the genetic diversity and availability of resources that enable systems genetic approaches in these strains.

Poster Number: 48

Uncovering Novel Functions Of Noradrenergic Locus Coeruleus Neurons

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Norepinephrine-synthesizing neurons of the locus coeruleus (LC) play a pivotal role in the regulation of stress. In vivo recording studies show that LC neuronal activity is evoked by stress, but suppressed by binge eating. Pharmacological studies in humans and rodents suggest that LC neurons may have a direct causal role in the production of anxiety and stress eating. To test this hypothesis, we developed a genetic strategy to selectively and noninvasively drive LC activity in awake, behaving animals. We generated a dual Flp/Cre-responsive mouse line (RC::FL-hM3Dq) that targets a Gq-coupled hM3Dq DREADD (Designer Receptor Exclusively Activated by Designer Drug) to the soma and dendrites of recombinase-expressing cells. To gain access to LC neurons, we exploited the fact that these neurons share a history of En1 and Dbh expression. In mice heterozygous for En1Cre, DbhFlpo, and RC::FL-hM3D, we tested whether the levels of hM3Dq expressed were sufficient to drive neural activity upon delivery of clozapine-N-oxide (CNO). We performed whole-cell recordings of LC neurons from En1Cre; DbhFlpo; RC::FL-hM3Dq mice and littermate controls and found that bath application of CNO (10 uM) depolarized hM3Dq-expressing LC neurons from En1Cre; DbhFlpo; RC::FL-hM3Dq. In contrast, CNO had no effect on membrane potential of LC neurons from littermate mice. To test whether activation of LC neurons would evoke anxiety, we treated En1Cre; DbhFlpo; RC::FL-hM3Dq and littermate mice with CNO (1 mg/kg or 5 mg/kg, IP) or vehicle before testing in three assays: the elevated plus maze, light dark box, and open field test. In all assays, CNO dose-dependently evoked anxiety-like behavior in En1Cre; DbhFlpo; RC::FL-hM3Dq mice. As expected, CNO had no effect on the behavior of littermate controls. Taken together, these findings demonstrate that hyperactivation of the LC results in anxiety, which is consistent with results obtained using optogenetic LC stimulation. Our intersectional chemogenetic strategy offers the unprecedented ability to non-invasively activate norepinephrine neurons of the LC during complex behaviors and provides the opportunity to discover new functions encoded by these cells. Experiments are underway to determine whether activation of the LC will directly promote maladaptive feeding (e.g., promote hypophagia, irregular eating patterns, or worsen stress-induced eating).

Poster Number: 49

Monitoring Subsecond Glucose Dynamics In Response To Intravenous Glucose And Cocaine Reveals Spatially Heterogeneous Microenvironments In The Rat Dorsal Striatum

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Brain cells utilize glucose to fuel metabolic processes, and glucose consumption is increased upon neuronal activation. Indeed, brain energy demands are high and this accounts for at least 20% of the entire body's glucose consumption. As such, real-time molecular detection of glucose dynamics is imperative to understanding the regulation of brain energy utilization and its involvement in neuropathological disorders, as well as the adaptations that occur upon exposure to substances of abuse. Extracellular glucose dynamics are dependent on two opposing forces: glucose availability (through cerebral blood flow) and glucose utilization. The location of intraparenchymal microvessels is heterogeneous across brain locations, and brain regions are differentially activated in response to stimuli. Therefore, it is likely that there is a great deal of variation in glucose dynamics both between brain regions, and within a single brain nucleus. In the caudate putamen (CPu), dopamine-related neurochemical adaptations occur in in habit-formation, goal-directed behaviors, and motor control. However, far less is known regarding glucose signaling in this brain region. Direct attempts to assess glucose heterogeneity within discrete brain locations have been hindered due to lack of technology available for these measurements with sufficient spatiotemporal resolution. This work employs fast-scan cyclic voltammetry (FSCV) in conjunction with glucose-oxidase (GOx) modified carbon fiber microelectrodes to monitor glucose dynamics with sub-second temporal resolution in the CPu. We have assessed heterogeneity in glucose signaling that occurs in response to intravenous administration of saline, glucose, and a cocktail comprised of cocaine and raclopride. These investigations will advance our understanding of the neuroenergetics at work in normal brain functions, as well adaptations associated with substance abuse.

Poster Number: 50

Unraveling the inhibitory synapse proteome in vivo.

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Inhibitory synaptic abnormalities result in multiple neurodevelopmental disorders that emerge in childhood, including autism spectrum disorders (ASD), epilepsy, neonatal hyperekplexia, and intellectual disability (ID). Like their excitatory counterparts, inhibitory synaptic properties are dynamic- their density, size, and receptor content is plastic during development and in response to activity. However compared to the excitatory synapse, the molecular components of the inhibitory synapse are not well defined, since this synaptic structure is not easily purified. This has impeded the systematic identification and analysis of the mechanisms underlying inhibitory synaptic function. To label, purify and identify the components of the postsynaptic inhibitory synapse, we developed a new approach to biotinylate proteins within this subcellular structure by expressing a biotin ligase (BirA) fused to Gephyrin (BirA-Gephyrin) in neurons within mice. BirA-Gephyrin showed punctate localization within the neuronal soma and dendritic shaft that co-localized with VGAT (a marker of inhibitory synapses). Biotinylated proteins also co-localized with BirA-Gephyrin. Purification of biotinylated proteins from mice expressing BirA-Gephyrin leads to an enrichment of multiple proteins, including some known to be components of the inhibitory synapse. Proteins that were not known to target inhibitory synapses were characterized by biochemical, immunostaining, and electrophysiological approaches in neurons. One of the novel proteins, InSyn1, interacts with Gephyrin and is localized to the inhibitory synapse in hippocampal neurons. Targeting of InSyn1 in single neurons with the CRISPR/Cas9 system, results in a reduction in mIPSC frequency, suggesting InSyn1 is critical for functional inhibitory synapses. These data validate the feasibility of a novel proteomic approach to target synaptic structures and reveal new proteins important for the molecular machinery of the inhibitory synapse.

Poster Number: 51

Exploring The Role Of Snca 3'utr In Parkinson's And Dementia With Lewy Body

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Synucleinopathies are a group of neurodegenerative diseases defined by the presence of intracellular aggregates, the Lewy Bodies (LBs), mainly composed of α-synuclein protein. This study focuses on two diseases in this group: Parkinson Disease (PD) and Dementia with Lewy Bodies (DLB). Both pathologies are characterized by α-synuclein aggregates in the neurons, however they have distinct characteristics with respect to the cell type containing the aggregates and the predominantly affected brain region. Specifically, the dopaminergic neurons are primarily affected in PD whereas DLB is predominantly defined by aggregates in the cholinergic neurons. Genome Wide Association Studies have implicated α-synuclein gene (SNCA) in the etiology of these diseases. Interestingly, genetic variability at the 3' region of SNCA locus has been associated with the risk to develop PD, while the 5' SNCA

region was implicated in DLB. However, the causal variants in each region of the SNCA locus and their mechanism of actions are still unknown. It has been suggested that α-synuclein levels are critical for the development of PD, and miRNA are known to regulate gene expression via interactions with UTRs. Here we investigated the role of SNCA 3' UTR in the regulation of SNCA expression in relation to the etiology of PD compared to DLB. A computational analysis using TargetScan identified four miRNA binding sites in SNCA 3'UTR. Herein, we studied the expression of these miRNAs in dopaminergic and cholinergic neurons differentiated from human Pluripotent Stem Cells. The miRNA expression levels have been evaluated at various stages of the differentiation. The dopaminergic and the cholinergic neurons show a distinctive pattern of the miRNAs at each stage of the differentiation; suggesting cell-specific regulation of the SNCA expression levels mediated by miRNAs. These results may explain the distinct molecular mechanisms underlying PD and DLB. Next, we examined the 3'UTR in a cohort consisted of individuals with three autopsy-confirmed neuropathological diagnoses, PD, DLB and normal controls. The SNCA 3' UTR was sequenced and several known and novel common genetic variants were identified. However, none of these genetic variants mapped within the predicted target sequence of the studied miRNAs. In conclusion, our data suggested that miRNAs participate in neuronal-type specific regulation of SNCA levels. Furthermore, common genetic variations may not contribute to the differential regulation of SNCA expression mediated by miRNAs, while, the role of rare variants warrants further investigations in larger cohorts.

Poster Number: 52

Voluntary Exercise Prevents Adolescent Binge Ethanol-Induced Serotonergic System Pathology In The Young Adult Raphe Nucleus

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Serotonergic neurons of the raphe nucleus regulate sleep, mood, endocrine function, and other processes that mature during adolescence. Alcohol abuse and binge drinking are common during human adolescence. We hypothesized that adolescent intermittent binge ethanol exposure would alter the developing serotonergic system leading to changes in adulthood. Using a Wistar rat model of adolescent intermittent ethanol (AIE; 5.0 g/kg, i.g., 2-day on/2-day off from postnatal day [P]25 to P55), we found a loss of dorsal raphe nucleus (DRN) serotonin (5-HT)-immunoreactive (+IR) neurons in late adolescence (P56) that persisted into adulthood (P220). Hypothalamic and amygdalar DRN serotonergic projections were reduced following AIE. Tryptophan hydroxylase 2, the rate-limiting 5-HT synthesizing enzyme, and vesicular monoamine transporter 2, which packages 5-HT into synaptic vesicles, were also reduced in the young adult midbrain following AIE treatment. Adolescent intermittent ethanol treatment increased expression of phosphorylated NF-κB p65 as well as markers of microglial activation (i.e., Iba-1 and CD11b) in the adult DRN. Administration of lipopolysaccharide to mimic AIE-induced microglial activation reduced 5-HT+IR and increased phosphorylated NF-κB p65+IR similar to AIE treatment. Voluntary exercise during AIE and across young adulthood prevented the AIE-induced loss of 5-HT+IR neurons, and blunted microglial marker and phosphorylated NF-κB p65+IR expression in the DRN. Together, these data report that AIE reduces serotonergic neurons in the adult brain, possibly through an innate immune mechanism, which might impact adult cognition, arousal, or reward sensitivity. Further, exercise prevents the deleterious effects of AIE on the serotonergic system of the young adult DRN.

Poster Number: 53

Glutamate Receptor Influence On Localized Oxygen Metabolism

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Functional hyperemia is the biological mechanism of maintaining homeostasis in the brain through mediating local cerebral blood flow (CBF) supply based on neuronal activity. According to hyperemia dogma, as neurons fire, CBF increases in a biphasic manner to provide blood rich with oxygen and glucose and renew locally depleted energy sources. Dysregulation of this system is noted in many disease states, including Alzheimer's and stroke. The relationship between CBF and neurotransmission important to understand, as healthy brains can experience decoupling between neuronal activity and blood flow in certain circumstances. Glutamate is the primary excitatory neurotransmitter that elicits cell firing within the brain. Its behavior is best characterized in the cortex or in slices, but technical issues have limited its study subcortically. We aim to better understand glutamatergic neurovascular influence deeper within an intact brain.
This research explores the role of glutamate on highly localized metabolic activity through simultaneous detection of cell firing and oxygen in response to glutamate. Iontophoresis, a local drug delivery technique, ejects glutamate and other drugs in close proximity to a neuron as opposed to less precise, systemic injections. Cell firing is detected using single unit recording electrophysiology and oxygen responses are recorded with fast-scan cyclic voltammetry, both at a single carbon fiber microelectrode. Our method of glutamate delivery is gentle enough to not provoke CBF changes, and instead probes the intensity and duration of increased oxygen metabolism demands that follow action potentials. We present glutamate-elicited

neuronal activation and subsequent metabolic changes as cells are exposed to a number of glutamatergic receptor antagonists in both the nucleus accumbens and somatosensory cortex

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Poster Number: 54

Real-Time Striatal Measurements of Oxidative Stress and Dopamine in the Dyskinetic Rat During Chronic L-DOPA Treatment for Parkinson's Disease

Leslie Wilson

Parkinson's disease (PD) is a chronic neurodegenerative disorder characterized by the preferential loss of dopaminergic neurons stemming from the substantia nigra pars compacta and innervating the dorsal striatum. The substantial decreases in striatal dopamine (DA) result in devastating hypokinetic movements and motor disturbances. Increased generation of reactive oxygen species, such as hydrogen peroxide (H2O2), is also thought to contribute to Parkinsonian symptoms. However, the precise role of H2O2 in the initiation, progression, and maintenance of the disease remains unclear, as reactive oxygen species are difficult to monitor in brain tissue. Further, several lines of evidence suggest that the standard treatment strategy of dopaminergic replacement therapy via administration of Levodopa (L-DOPA; L-3,4 dihydroxyphenylalanine) may serve to increase oxidative stress and potentiate cell death. We aim to investigate how striatal H2O2 and DA dynamics underlie behavioral changes that result from chronic L-DOPA administration in a rodent model of PD (unilateral 6-OHDA lesion) using fast-scan cyclic voltammetry, an electrochemical technique that affords precise spatial and temporal resolution, as well as selective detection of these neurochemicals. Specifically, carbon-fiber microelectrodes are used to simultaneously quantify rapid H2O2 and DA fluctuations at single recording sites in the dorsal striatum over several weeks of L-DOPA administration. The chemical fluctuations are correlated with behavioral abnormalities that develop over the course of treatment. These studies will aid in our understanding of how oxidative stress modulates nigrostriatal DA signaling, and will demonstrate how these signals correspond with the development of dyskinetic movements in the treatment of PD.

Poster Number: 55

Role of Hydrogen Peroxide Fluctuations in the Nucleus Accumbens During Cocaine-induced Locomotor Activity

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Behavioral sensitization is the augmented motor-stimulant response that occurs with repeated, intermittent exposure to most drugs of abuse, including cocaine. Much research has been conducted to determine the neural mechanisms of sensitization, which includes the neuroadaptations of the mesolimbic dopamine system. Acute cocaine exposure is well known to induce increases in extracellular dopamine level in the nucleus accumbens (NAc), which receives dopamine input from the ventral tegmental area (VTA). Given that recent study has shown that hydrogen peroxide (H2O2) can inhibit dopamine release in the striatum, the present study intends to examine the fluctuations of H2O2 and dopamine simultaneously in the NAc during cocaine-induced locomotor activity. To this end, we implanted chronic microelectrode in the NAc of Sprague-Dawley rats. After rats were recovered from surgery, we measured H2O2 and dopamine concentrations in the NAc of rats and recorded rat locomotor activity simultaneously after acute cocaine (15 mg/kg i.p.) or saline injections. We found that H2O2 concentration in the NAc was not altered during rat movements after saline injection, as compared rats resting periods. However, H2O2 concentration in the NAc was increased during rat movements, but decreased during resting periods after acute cocaine injection. Reversely, dopamine concentrations in the NAc was attenuated during rat movement, but enhanced during resting periods after acute cocaine injection. These results suggested that dopamine concentration in the NAc fluctuates during locomotor activity after acute cocaine exposure. It is likely that acute cocaine-induced release of H2O2 negatively regulates dopamine levels in the NAc. This line of study will help shed new light on the motor-stimulant response of cocaine, and have the potential to provide novel insight into the mechanisms of cocaine-induced sensitization.

Poster Number: 56

Cannabinoids Occlude The Hiv-1 Tat-Induced Decrease In Gabaergic Neurotransmission In Medial Prefrontal Cortex Slices

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In the era of combined antiretroviral therapy (cART), human immunodeficiency virus type 1 (HIV-1) is now considered a chronic disease that specifically targets the brain and causes HIV-associated neurocognitive disorders (HAND). Endocannabinoids exhibit neuroprotective and anti-inflammatory properties in several central nervous system (CNS) disease models, but their effects in HAND are poorly understood. To address this issue, whole-cell recordings were performed on young (14 – 21 day old) C57BL/6J mice. We investigated the actions of the synthetic cannabinoid WIN55,212-2 (1 μM) and the endocannabinoid N-

arachidonoyl ethanolamine (anandamide; AEA, 1 μM) in the presence of HIV-1 Tat on GABAergic neurotransmission in mouse prefrontal cortex (PFC) slices. We found a Tat concentration dependent (5 – 50 nM) decrease in the frequency and amplitude of miniature inhibitory postsynaptic currents (mIPSCs). The cannabinoid 1 receptor (CB1R) antagonist rimonabant (1 μM) and zero extracellular calcium prevented the significant Tat-induced decrease in mIPSCs. Further, bath-applied WIN55,212-2 or AEA by itself, significantly decreased the frequency, but not amplitude of mIPSCs and/or spontaneous IPSCs (sIPSCs), and occluded a further down-regulation of IPSCs by Tat. Pretreatment with rimonabant (1 μM) but not the CB2R antagonist AM630 (1 μM) prevented the AEA-induced decrease in IPSCs frequency without any further Tat effect. Results indicated a Tat-induced decrease in GABAergic neurotransmission, which was occluded by cannabinoids via a CB1R-related mechanism. Understanding the relationship between Tat toxicity and endocannabinoid signaling has the potential to identify novel therapeutic interventions to benefit individuals suffering from HAND and other cognitive impairments.

Poster Number: 57

Closed loop dorsal column stimulation for epilepsy control

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Although electrical neurostimulation has been proposed as an alternative treatment for drug-resistant cases of epilepsy, current procedures such as deep brain stimulation, vagus, and trigeminal nerve stimulation are effective only in a fraction of the patients. Here we demonstrate a closed loop brain-machine interface that delivers dorsal column electrical stimulation (DCS) as a way to suppress epileptic seizures. Rats were implanted with cortical recording microelectrodes and planar spinal cord stimulating electrodes, and then injected with pentylenetetrazole to induce seizures. Seizures were detected in real time from cortical local field potentials, after which DCS was applied. This method decreased seizure episode frequency by 44% and seizure duration by 38%. We argue that the therapeutic effect of DCS is related to modulation of cortical theta waves, and propose that this closed-loop interface has the potential to become an effective and minimally invasive treatment for refractory epilepsy and other neurological disorders.

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Notes


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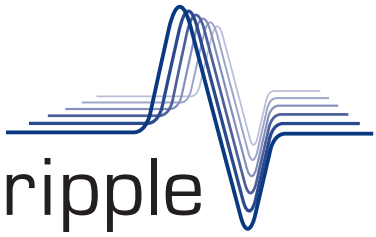
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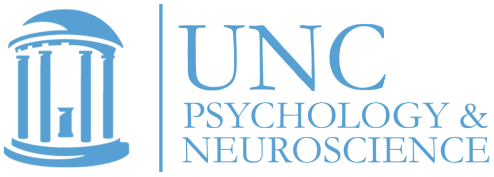
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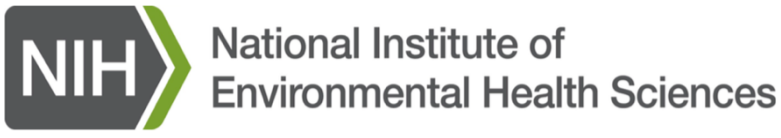
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