UCONN HEALTH

Ray A. Kroc and Robert L. Kroc
DEPARTMENT OF NEUROSCIENCE
ANNUAL RETREAT

PROGRAM & AGENDA

Thursday, May 10, 2018
8:00 am – 6:00 pm

The Mark Twain House & Museum

351 Farmington Avenue

Hartford, CT 06105

Stephen J. Crocker, Ph.D
Associate Professor, Department of Neuroscience
Associate Director, Neuroscience Graduate Program
University of Connecticut Health Center
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10 May 2018

Dear Neuroscience Program Faculty, Postdocs, Students and Guests,

Welcome to the 17th annual Neuroscience Program Retreat!

This year's event is again being hosted at the Mark Twain House in Hartford. One of the best known American writers, Mark Twain was a remarkable man and the Mark Twain House is recognized as a National Historic Landmark.

Complementary tours of the house will be made available to all participants, please sign-up when you check-in on the morning of the event. We hope you will take advantage of this tremendous opportunity to visit one of the best places in Hartford!

There are a few housekeeping items we would like to bring to your attention. **First, you MUST BRING YOUR UNIVERSITY I.D.** This will allow others to know who you are, it will allow the Mark Twain House staff identify you as part of our event, and lastly it will also allow you 10% off in the Mark Twain gift shop! Second, the venue is air conditioned. This means that the temperature can vary quite a bit. If you have a tendency to get cold, please remember to bring a sweater or light jacket. Third, while parking is free, it is not unlimited. Please make every attempt to carpool so that we may minimize the total number of cars at the Mark Twain house. Driving instructions are provided in the pages following this letter.

Posters will be presented in two sessions. If you are a poster presenter, please check which session you have been assigned before putting up your poster. Session 1 will be before lunch, Session 2 immediately after. If you are giving a poster, the page number on you abstract in this book will be the number of poster board upon which you should put your poster.

Oral presentations will be made in the "Auditorium". Every speaker must upload their talk before the beginning of their session. There will be a computer available and this is the only one presenters may use. No personal computers will be allowed to be used for presentations because the system at the Mark Twain house requires rebooting if the computers are changed and switching from computer-to-computer also adds time to an already tight schedule. All presenters are strongly encouraged to upload and test their presentations on the laptop which will be made available Thursday in the Departmental Office. Thanks in advance for your cooperation and understanding.

Please print or carry an electronic copy of this program with you to the meeting. Printed copies will not be provided for all participants at this year's event. Please also know that there will be free wireless internet access complements of the Mark Twain House. The wifi

password will be provided at the meeting.

Presentations by students and postdocs will be judged for awards. Judges may or may not identify themselves as judges. You will be judged on a variety of criteria including, but not limited to: knowledge of your subject matter, the quality of your presentation and your responses to questions. Winners will be announced immediately before the closing remarks.

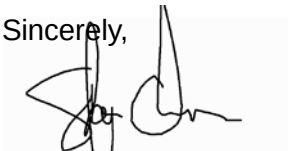
The retreat will wrap-up with a keynote address and reception to follow.

If you have any suggestions for improving the event for next year, please don't hesitate to let either myself or Jody Gridley know as we begin organizing this event for next year in only a couple of months time.

We hope you enjoy the day and find your experience helpful and rewarding.

On behalf of the Organizing Committee,

Sincerely,



Stephen J. Crocker, Ph.D.
2018 Neuroscience Retreat Organizer

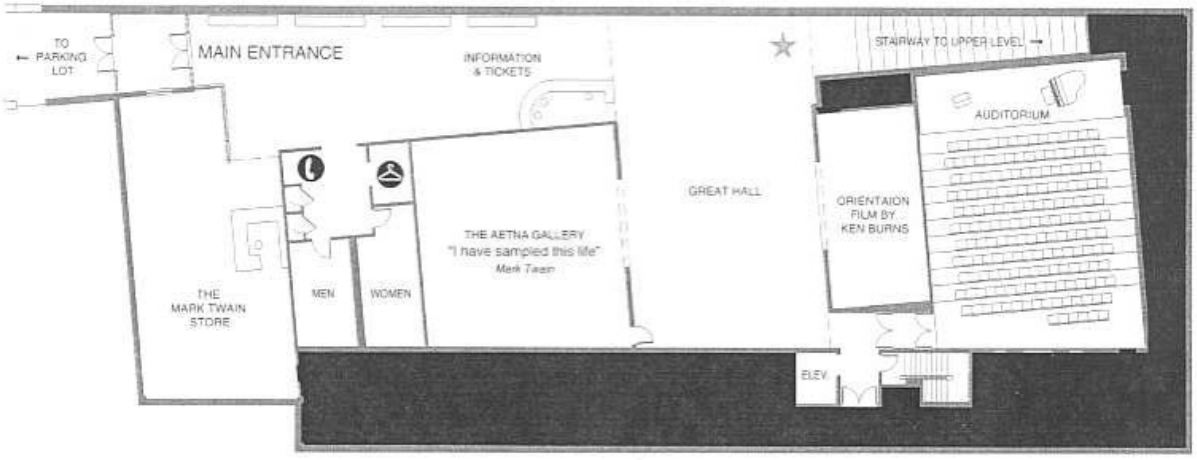
2018 Meeting Program

Time	Event	Location
8:00 – 8:45 am	Registration & Sign-in Continental Breakfast	Great Hall
8:45 - 9:00 am	**Welcome Address** Dr. Stephen Crocker	
9:00 - 9:45 am	**Symposium A**	Auditorium
9:45 - 10:00 am	Coffee Break	Great Hall
10:00 – 11:00 am	**Symposium B**	Auditorium
11:00 – 12:00 am	**Poster Session 1**	Great Hall
12:00 - 1:00 pm	**Lunch & Tours**	Great Hall
1:00 - 2:00 pm	**Poster Session 2**	Great Hall
2:00 - 2:15 pm	Vendor Recognition Presentation	Auditorium
2:15 - 3:15 pm	**Symposium C**	Auditorium
3:15 - 3:30 pm	Coffee Break	Great Hall
3:30– 4:30pm	**Keynote Address**: Junying Yuan, PhD Harvard University *"Role of RIPK1 in mediating neuroinflammation and cell death in human neurodegenerative diseases"*	Auditorium
4:30 pm	**Presentation of Awards** & Closing Remarks	
4:45 - 6:00 pm	Gelato – Social event	Outdoor Terrace*

*Weather permitting – alternate location: Great Hall

The Mark Twain House & Museum

Lower Level / Main Entrance



★ TOURS START HERE

Upper Level

↑ TO THE MARK TWAIN HOUSE

3rd Floor–Research Library (By Appointment Only)

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Driving Directions to:

The Mark Twain House & Museum
351 Farmington Avenue
Hartford, CT 06105

From Springfield and Points North

Take I-91 South to I-84 West in Hartford. Take Exit 46, Sisson Avenue. At the traffic light, turn right (north) onto Sisson. Continue four blocks to the end of Sisson Avenue. At the traffic light, turn right (east) onto Farmington Avenue. Continue three blocks (approx. 2/10 mile) on Farmington and look for the sign on the right at the entrance to the museum's free parking lot. ***Note:*** The entrance to the parking lot is one block before The Mark Twain House.

From New York City, New Haven and Points South

Take I-91 North to I-84 West in Hartford. Take Exit 46, Sisson Avenue. At the traffic light, turn right (north) onto Sisson. Continue four blocks to the end of Sisson Avenue. At the traffic light, turn right (east) onto Farmington Avenue. Continue three blocks (approx. 2/10 mile) on Farmington and look for the sign on the right at the entrance to the museum's free parking lot. ***Note:*** The entrance to the parking lot is one block before The Mark Twain House.

From Waterbury and Points West

Take I-84 East to Hartford; take Exit 46, Sisson Avenue. At the traffic light, turn right onto Sisson. Continue four blocks to the end of Sisson Avenue. At the traffic light, turn right (east) onto Farmington Avenue. Continue three blocks (approx. 2/10 mile) on Farmington and look for the sign on the right at the entrance to the museum's free parking lot. ***Note:*** The entrance to the parking lot is one block before The Mark Twain House.

From Boston and Points East

Take I-84 West to Hartford; take Exit 46, Sisson Avenue. At the traffic light, turn right onto Sisson. Continue four blocks to the end of Sisson Avenue. At the traffic light, turn right (east) onto Farmington Avenue. Continue three blocks (approx. 2/10 mile) on Farmington and look for the sign on the right at the entrance to the museum's free parking lot. Note: The entrance to the parking lot is one block before The Mark Twain House.

For Passengers with Limited Mobility or LOADING DOCK

(65 Forest Street Entrance)

Follow the above directions, but continue east on Farmington Avenue and go past The Mark Twain House. Take your first right onto Forest Street and look for the sign on the right for the entrance to the Museum Center's rear parking lot.

KEYNOTE SPEAKER

“Role of RIPK1 in mediating neuroinflammation and cell death in human neurodegenerative diseases”

Junying Yuan, PhD

Elizabeth D. Hay Professor of Cell Biology
Harvard Medical School, Boston, MA

Junying Yuan made critical contributions to our understanding of apoptosis and necroptosis, two fundamental mechanisms that regulate the survival and death of mammalian cells. Her Ph.D. work, as a student at the Harvard Medical School and conducted in the laboratory of H. Robert Horvitz at the Massachusetts Institute of Technology, provided the first insight into the mechanism of programmed cell death in the nematode *C. elegans*. Yuan started her own lab in 1990 at the Cardiovascular Research Center, Massachusetts General Hospital, to test her hypothesis that a similar programmed cell death mechanism might exist in mammalian cells. Her daring hypothesis was proved two years later when her laboratory demonstrated that the mammalian interleukin-1b converting enzyme (later named caspase-1) is a functional homologue of *C. elegans* cell death gene product Ced-3 (Miura et al. 1993) and inhibition of caspase activation blocks neuronal cell death induced by trophic factor deprivation (Gagliardini et al. 1994). These works provided the first insight into the molecular mechanism that regulates apoptosis in mammalian cells. Subsequently, after her move in 1996 to Department of Cell Biology, Harvard Medical School, Yuan lab discovered necroptosis, a regulated necrotic cell death mechanism, and the role of RIPK1 kinase as a key mediator of necroptosis (Degterev et al. 2005, Degterev et al. 2008). This discovery overturned the traditional dogma that necrosis can only be a form of unregulated passive cell death and demonstrated the possibility of inhibiting necrotic cell death in multiple forms of degenerative and inflammatory human diseases. A small molecule inhibitor of RIPK1 kinase (Nec-1) discovered by Yuan lab is currently in preparation for a human clinical trial targeting neurodegenerative disease.

Symposium A: Talk No. 1

Recruitment of neurons into neural ensembles based on dendritic plateau potentials

Peng P. Gao[1*], Joe W Graham[2], Sergio L Angulo[2], Salvador Dura-Bernal[2], Michael L Hines[3], William W Lytton[2], Srdjan D Antic[1]

[1]Neuroscience Department, University of Connecticut Health Center, Farmington CT 06030, USA
[2]Department Physiology & Pharmacology, SUNY Downstate, Brooklyn, NY 11203, USA
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Experimental observations have shown that glutamatergic inputs to the basal dendrites of cortical pyramidal neurons activate AMPA and NMDA receptors, which can bring the dendrites into a long-lasting depolarized state: a dendritic plateau potential. These sustained depolarizations push the cell body towards spike threshold and reduce the membrane time constant. In such a "Prepared" state, the pyramidal cells can respond to other sparse synaptic inputs more quickly and easily, facilitating synchronization of firing. During the plateau depolarization, a neuron can tune into ongoing network activity and synchronize spiking with other neurons to provide a coordinated “Active” state (robust firing of somatic action potentials), which would permit "binding" of signals through coordination of neural activity across a population. Under this scenario, Active cells are recruited from cells in the Prepared state, and therefore the transient Active ensemble is embedded in the longer-lasting Prepared ensemble of neurons. We hypothesize that "embedded ensemble encoding" may be an important organizing principle in networks of neurons, explaining how electrical signaling endows central nervous system with capacity to form large number of neural ensembles. We have developed a morphologically-detailed model reconstructed from a cortical Layer 5 prefrontal pyramidal cell in the NEURON simulator. Both synaptic AMPA/NMDA and extrasynaptic NMDA receptors are placed on basal dendrites to model the induction of plateau potentials. The active properties of the cell are tuned to match the amplitude and duration of plateau potentials recorded by voltage-sensitive dye imaging in dendrites and whole-cell patch measurements in soma. Then, the effects of input location, receptor conductance, calcium-activated potassium channels and voltage-activated calcium channels were explored in the model. These findings help us to better understand the implications of dendritic plateaus at the cellular and network level. In the future, this detailed individual cell model can be used to develop cortical meso-scale network models for exploring the hypotheses pertaining to the recruitment of neurons into neural ensembles.

Supported by NIH Brain Initiative R01 EB022903, U01EB017695

Symposium A: Talk No. 2

Voltage and calcium signals in dendrites of medium spiny neurons.

Jinyoung Jang and Srdjan D. Antic

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The medium spiny neuron (MSN) is a key neuron in the basal ganglia circuit, involved in several major neurological disorders including Parkinson's disease, Huntington's disease and attention deficit hyperactivity disorder (ADHD). MSNs exhibit two neural states. The "Down state" (hyperpolarized membrane potential) alternates with the "UP state" (depolarized membrane potential) in accordance to the temporal availability of glutamatergic synaptic inputs impinging onto their spiny dendrites. Therefore, to understand the functional roles of MSNs, it is necessary to study dendritic physiological properties. Intrinsic membrane properties of MSN dendrites are poorly understood because thin dendritic branches do not tolerate glass electrode recordings. Here, we performed simultaneous somatic whole-cell recording with dendritic imaging; either voltage sensitive dyes or Ca^{2+} indicators. Dendritic membrane potential changes were induced by backpropagating action potentials (bAPs) or local application of glutamate on dendrite. Our results reveal an increasing dendritic AP peak latency depending on the distance from the soma. The amplitudes of action potential-induced dendritic Ca^{2+} influxes decrease with distance from the soma suggesting a distance dependent attenuation of backpropagating action potentials in dendrites of MSNs. Next, we characterized the voltage waveforms of glutamate evoked plateau potentials in dendrite and cell body simultaneously. Invariably, the dendritic plateaus preceded somatic plateau potentials and the initiation phase of the dendritic UP state (plateau) was steeper than that in the soma, consistent with a local initiation of the dendritic plateau. Detailed characterization of voltage and Ca^{2+} signals is expected to reveal the intrinsic dendritic properties and bring us closer to the functional roles of MSNs in the basal ganglia circuits.

Supported by NIH Grant: U01MH109091

Symposium A: Talk No. 3

The constitution of dystrophic neurites in aging and in Alzheimer's disease mouse model brain and their 3D structures

Md. Golam Sharoar, Xiang-You Hu and Riqiang Yan

Department of Neurosciences, UCONN Health, Farmington, CT 060032

Formation of dystrophic neurites (DNs) is one of the major pathological features and a contributing factor of synaptic dysfunction in Alzheimer's disease (AD) patient's brain. We found that reticulon 3 (RTN3), a protein known to be enriched in the tubular endoplasmic reticulum, plays a role in forming RTN3 immunoreactive DNs (RIDNs), both in aging and in AD brains. Our recent study showed that RIDNs also contain other tubular ER shaping proteins such as REEP2 and REEP5, and ER tubules are abnormally accumulated in the RIDNs where mitochondria were also degenerated. In this study, we aimed to understand how RIDNs are developed in AD mice (5xFAD and APP/PS1ΔE9 mice) and how they are related to or differed from DNs that are enriched with other proteins. We observed that proteins such as RTN3, ubiquitin and ATG9A are enriched in DNs in different time points when comparing to the formation of amyloid plaques. We identified that several autophagy proteins and a late endosomal protein are accumulated only in plaque- associated DNs, and they were partially co-localized with RIDNs. An early autophagy protein, ATG9A, was found to constitute DNs during very initial stage of plaque development, when RIDNs were also started to form. We employed 3D electron microscopic approach to reconstruct the structural morphology of different types of DNs including RIDNs, which are predominantly enriched by tubular ER accumulation in axons, and DNs that are mostly enriched by multi-vesicular bodies. We conclude that RIDNs are constitutes predominantly by dysfunctional tubular ER accumulation during aging, while deposition of amyloid beta could induce clustered RIDNs and DNs by impairing autophagy, ubiquitin proteasome system and normal ER distribution in the axons those appeared as a diverse morphology surrounding an amyloid plaque.

Presenting Author: Md. Golam Sharoar
Email: sharoar@uchc.edu

Funding Source: NIH

Symposium B: Talk No. 4

The Effect of γ-Synuclein Treatment on Human Cortical Astrocyte Proliferation and BDNF Expression

Cynthia Winham (Winham@Hartford.edu) and Andrew Koob, Ph.D.

University of Hartford, M.S. Neuroscience Program

Abstract: γ-Synunclein (γ-syn), a member of the synuclein protein family implicated in neurodegenerative disease, is expressed in astrocytes in the human nervous system, and increased extracellularly in the brain and cerebrospinal fluid of individuals diagnosed with Alzheimer's disease. Upregulation of γ-syn has also been shown in glioblastomas and other cancers. The purpose of this study is to better understand the function of extracellular γ-syn, through an examination of the relationship between γ-syn treatment and cell proliferation in human cortical astrocytes. Astrocytes from adult human cortex were cultured *in vitro* and treated with 100 nM γ-syn for 3, 6 and 24 hours. Western blot analysis revealed endogenous expression of γ-syn in controls as well as uptake of γ-syn by human cortical astrocytes at all time points. Brain derived neurotophic factor (BDNF), a growth factor released by astrocytes, was reduced intracellularly after 6 hours of treatment compared to controls, which coincided with elevated extracellular BDNF levels. Then after 24 hours of treatment, extracellular BDNF was decreased compared to controls. Immunocytochemistry of proliferation marker 5-bromodeoxyuridine (BrdU) after γ-syn treatment was increased at 3 and 6 hours, whereas BrdU and Ki67 labeling was not significant by 24 hours compared to controls. Additionally, after cell synchronization in G_0/G_1, cells were released back into the cell cycle and treated with γ-syn. Analysis of BrdU and propidium iodide through flow cytometry 24 hours after release revealed an increase in G_2/M phase of the cell cycle in cells treated with100 nM γ-syn compared to controls. This effect was no longer seen at 48 hours. These results suggest γ-syn treatment initially causes upregulation of the cell cycle and BDNF release in human cortical astrocytes. Since stimulation of reentry into the cell cycle by non-neuronal cells is shown in neurodegenerative disease, elevated levels of extracellular γ-syn could potentially contribute to this process.

Support: Support for this work was provided in part by The Women's Advancement Initiative, advancing each woman's potential is the HCW tradition of the University of Hartford. Funding was also provided by University of Hartford College of Arts and Sciences Dean's Office and Neuroscience Program.

Symposium B: Talk No. 5

Exosomes induce relapsing-remitting disease in a MOG-EAE model of demyelination

Cory M. Willis1, Alexandra M. Nicaise1, Andrew Mendiola2,3, Antoine Menoret4, Evan R. Jellison4, Katerina Akassoglou2,3, Anthony T. Vella4, and Stephen J. Crocker1,5

Departments of Neuroscience1 and Immunology4, University of Connecticut School of Medicine, Farmington, CT
Gladstone Institute of Neurological Disease2 and Department of Neurology3, University of California, San Francisco, CA

Extracellular vesicles (EVs) represent a conserved primordial form of intercellular communication. Accumulating evidence now support exosomes, a 50-150 nm class of EVs, as important signaling mediators in health and disease that may contribute to development of pathology in the central nervous system (CNS). In this study, we investigated the role of blood exosomes in development of demyelinating autoimmune disease. Donor exosomes were isolated from whole blood plasma of naive C57Bl/6 mice (6-12 weeks of age) and then intravenously administered to C57Bl/6 mice that had been immunized with myelin oligodendrocyte glycoprotein (MOG35-55) at the time of peak clinical illness in this model of experimental autoimmune encephalomyelitis (EAE). The course of clinical disease in non-exosome-inoculated EAE mice was consistent with the monophasic profile of ascending paralysis beginning around day 12 and abating by day 21. In contrast, all exosome-treated EAE mice developed multiple spontaneous clinical relapses through the 51 day experimental period. While MOG-EAE is known to evoke strong CD4+ T cell responses, CyTOF analysis of infiltrating leukocytes into the spinal cord of exosome-inoculated EAE mice determined that these clinical relapses were driven by CD8+ T cells. Administration of anti-CD8 antisera was found to effectively block these relapsing-remitting episodes induced by exosome innoculation. High through-put proteomic analysis (PF-2D) of exosomes followed by tandem mass spectrometry identified fibrinogen alpha-chain (FGA) as a key component in exosomes, which we surmised was a driver of the clinical phenotype in EAE mice. Isolation of exosomes from the blood of naive FGA knockout mice when inoculated into MOG-EAE mice was not found to evoke any exacerbations of disease or clinical relapses, while blood exosomes isolated from wildtype FGA littermate controls were found to recapitulated the relapse-remitting clinical phenotype in EAE mice. These findings highlight a potentially novel connection between the role of exosomes as a source of the blood coagulant factor FGA to the development of the prominent CD8+ T cell-driven immunopathology which is observed in multiple sclerosis patients.

This work was supported by the National Institutes of Health (R21-NS087578-01A1 to SC and AT) and R56 (NS-NS099359-01A1 to SC). We acknowledge the W.M. Keck Foundation Biotechnology Resource Laboratory at Yale University for mass spectrometry analysis.

Symposium B: Talk No. 6

Tissue Inhibitor of Matrix Metalloproteinase-1 prevents the development of hypersensitivity following cutaneous inflammation.

Knight BE1, 2, Yasko JR1, 2, Kozlowski N.3, Crocker SJ.1, King T.6, Young EE.2, 3, 4, 5, Baumbauer KM.1, 2, 3, 5

1Department of Neuroscience, University of Connecticut Health Center, Farmington, USA 2Center for Advancements in Managing Pain, University of Connecticut, Storrs, USA 3The Center for Advancement in Managing Pain, School of Nursing, University of Connecticut, Storrs, USA 4Genetics and Genome Sciences, University of Connecticut Health Center, Farmington, USA 5Institute for Systems Genomics, University of Connecticut Health Center, Farmington, USA 6University of New England, Biddeford, Maine, USA

Chronic pain is a significant health concern that affects millions of people worldwide. Persistent inflammation is known to contribute to the development of chronic pain, and thus, identifying molecules that regulate the inflammatory process and aid in its normal resolution may aid in the development of novel treatments. Peripheral injury and inflammation result in the dynamic release of molecules that aid in tissue repair as well as result in hypersensitivity. Research has shown that proteases important for tissue remodeling, matrix metalloproteinase-9 and its endogenous inhibitor, tissue inhibitor of matrix metalloproteinase-1 (TIMP-1) contribute to the development of sensitivity following nerve damage. However, the specific role of TIMP-1 in development of inflammatory hypersensitivity is not well understood. We hypothesized that TIMP-1 expression aids in the prevention of hypersensitivity following inflammation. To test this, we subcutaneously (s.c.) injected complete Freund's adjuvant (CFA) into the right hind paw of wildtype mice. We then randomly assigned mice to behavioral assessment or for protein analysis at days 1, 3, 5, and 7 following CFA-injection. We found that cutaneous TIMP-1 expression was upregulated prior to the onset of mechanical hypersensitivity in WT mice. We also found that keratinocytes upregulate TIMP-1 protein following in vitro inflammation. To test whether TIMP-1 expression contributed to the development of inflammatory sensitivity we used a murine global knockout for TIMP-1 (T1KO). When we assessed mechanical sensitivity, and found that WT mice developed hypersensitivity 3-5 days following inflammation that resolved by day 7. Conversely, T1KO mice developed a rapid-onset hypersensitivity within 24 hr of inflammation that persisted for more than 7 days. To determine the sufficiency of TIMP-1 in the regulation of inflammatory hypersensitivity, we administered recombinant murine (rm)TIMP-1 (s.c.) at the time of CFA injection and found that rmTIMP-1 attenuated inflammatory responding. Interestingly, we did not detect any differences in the local inflammatory response between genotypes or the amount of MMP-9 activity, suggesting that TIMP-1 may prevent the onset of sensitivity through a non-traditional pathway. To test this, we administered N terminus MMP regulatory subdomain, C terminus trophic factor subdomain, or full-length TIMP-1 protein (s.c.) at the time of CFA injection and found that hypersensitivity was attenuated in all conditions. These data suggest cutaneous TIMP-1 expression during the onset of inflammation is important for preventing the development of sensitivity through both MMP regulation as well as trophic factor signaling mediated through TIMP-1's receptor, CD63. Previously it has been shown that TIMP-1/ CD63 signaling can target β-catenin for degradation and prevent neuropathic pain. We propose to investigate this pathway in our current model to understand how TIMP-1 may attenuate inflammatory sensitivity. Collectively, administration of TIMP-1 may be a potential therapy to prevent the development of clinical pain.

Funding: R03NS096454, UCONN/UCHC InCHIP Grant, UCHC Convergence Grant, Rita Allen Foundation

Symposium B: Talk No. 7

Bladder vs. Urinary Aging: A Multimode Correlation Study in Mice

Hardy, C.H.[1,2], Knight, B.E.[2], Al-Naggar, I.M.[1], Kuchel, G.A.[1,2], Smith, P.P.[1,2,3,4]

[1]Center on Aging, University of Connecticut School of Medicine, UConn Health
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[3]Department of Surgery, University of Connecticut School of Medicine, UConn Health
[4]Institute for Brain and Cognitive Science, University of Connecticut

With age, the prevalence of urologic dysfunction and lower urinary tract (LUT) symptoms increases. While symptomatic patients have been highly studied, little is known about what separates the dysfunctional elderly from the resilient population that maintains effective LUT function. In this on-going study, voiding behavior, urodynamic function, detrusor response, bladder architecture, and gene expression changes will be analyzed across four age groups (2-4 mo, 10-14 mo, 18-22 mo, and 24+ mo) and both sexes in WT C57/Bl6 mice. In this within-subjects design, we seek to correlate cystometric performance with changes in gene expression, voiding behavior, and/or structural changes that occur with aging, potentially providing insight into the underlying mechanisms determining dysfunction or resilience.

Symposium C: Talk No. 8

Age-related hearing loss – how a monoallelic single point mutation impairs auditory processing

A. Burghard, N. Morel, D. L. Oliver

The *Ahl* mutation in the Cdh23 gene ($Cdh23^{753A>G}$) is known to lead to the age-related hearing loss phenotype observed in many inbred mouse strains. This phenotype involves an early onset of age-related high frequency hearing loss, which leads to moderate hearing deficits at only 6-months of age, and severely hearing impaired animals within one year. While previous studies on Cdh23 mutations have focused on the comparison of homozygous for the mutation vs homozygous wild-type, we investigated how the *Ahl* mutation on a single allele alters auditory capabilities of mice over age. To compare auditory phenotypes to the specific genotype, animals were genotyped via Saenger sequencing of genomic DNA. The hearing thresholds were established using click-evoked auditory brainstem response (ABR) and amplitude-modulation following response (AMFR) measurements. The latter was also used to assess the temporal processing abilities of the animals. For the audiogram, the AMFR stimuli had a 0.3 octave narrow-band noise carrier frequency and a 42.9 Hz modulation frequency shaped by a sine wave raised to the exponent 8. To test the temporal processing abilities, the frequency with the lowest threshold was stimulated at 30 dB above threshold and the modulation frequency was varied from 17-544 Hz. Mice homozygous for the $Cdh23^{753A/A}$ mutation showed a high frequency hearing loss beginning at 3 months of age which progressed until no high frequency hearing was detectable at one year. In contrast, the $Cdh23^{753G/A}$ mice did not show hearing loss and showed no differences in hearing thresholds across ages or compared to $Cdh23^{753G/G}$ mice. However, other aspects of hearing function of heterozygous mice were affected. We looked at sound intensity coding via amplitude (amplitude growth function) at the level of the auditory nerve (ABR wave I). While it shows a steep growth for young animals, the curve flattens for the one-year old group in $Cdh23^{753A/A}$ and $Cdh23^{753A/G}$ mice, but not $Cdh23^{753G/G}$ mice. This indicates an intensity coding deficit in $Cdh23^{753A/A}$ and $Cdh23^{753A/G}$ mice. Moreover, when the temporal precision of the AMFR was tested, we found a decline in peak synchrony for high modulation frequencies in mice homozygous and heterozygous mice for the mutation. This also indicates a peripheral deficit. In conclusion, the *Ahl* ($Cdh23^{753A>G}$) mutation has effects on auditory abilities besides hearing thresholds. These deficits in temporal precision and intensity coding were also present in animals heterozygous for this mutation. Since Cdh23 mutations are seen in mouse and human, it is important to assess auditory processing and hearing in both heterozygous and homozygous individuals in more detail.

This study was supported by NIH R21DC013822 and UConn Health Research program (N.M.)

Symposium C: Talk No. 9

Transcriptional alterations in sensory neurons and behavioral assessments reveal impact of ASIC3 following Spinal Cord Injury

Jessica R. Yasko[1], Kaela Drzewiecki[4], Yashasvee Munshi[2], Justin Pranulis[5], Erin E. Young[3], and Kyle M. Baumbauer[1]
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While spinal cord injury (SCI) is typically associated with loss of locomotor function, SCI can also result in chronic pain, affecting nearly 67% of patients with SCI. Most research concerning mechanisms responsible for SCI pain emphasize alterations within the spinal cord. However, recent studies suggest that SCI and the subsequent injury-related processes also impact primary sensory neurons below the level of injury. To begin to address how peripheral sensory neurons respond to a centrally generated injury, we examined SCI-induced changes in gene expression of individual sensory neurons at two different injury severities, and the skin in which they innervate. Using single cell PCR to profile patterns of nociceptor gene expression, we found that expression of acid sensing ion channel 3 (ASIC3) mRNA is increased in L2 and L3 DRG neurons 24hrs following thoracic spinal contusion injury. Increased ASIC3 expression may also correlate with the development of spontaneous afferent firing, suggesting that ASIC3 could contribute to SCI-induced hypersensitivity. Because of this, we also examined mechanical (von Frey) and thermal (Hargreaves) behavioral hypersensitivity. Behavioral data demonstrate that mice administered the ASIC3 antagonist APETx2, or an antisense oligonucleotide against both isoforms of ASIC3, show reduced mechanical sensitivity following SCI. Ongoing experiments are addressing the mechanism in which ASIC3 is upregulated following injury. Collectively, our data suggest that ASIC3 may serve to detect the early stages of central injury in below-level afferents, which may also contribute to SCI-induced pain.

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Symposium C: Talk No. 10

Establishing a morphological phenotype for Angelman Syndrome and Chromosome 15q Duplication Syndrome using stem cell-derived neurons

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In mature neurons, *UBE3A*, which encodes for an ubiquitin ligase, is only expressed by the maternal allele due to genomic imprinting. Loss of the maternal copy of *UBE3A* results in Angelman Syndrome (AS), a neurodevelopmental disorder characterized by absent speech, severe seizures, intellectual disability, motor dysfunction and a happy disposition. On the other hand, the duplication of the maternal region surrounding *UBE3A* results in Chromosome 15q Duplication Syndrome (Dup15q). Dup15q is the most common chromosome abnormality associate with autism spectrum disorder, but other common symptoms include developmental and motor skill delays and seizures. Mouse models of AS indicate that there is a reduction of dendritic spines, while a common characteristic of autism is an increase of dendritic spines in both human and mice. However, it is still unclear how human neurons with these specific disease-causing mutations are affected. Here we use patient-specific induced pluripotent stem cells to examine the maturation of forebrain neurons in both of these disorders. We have developed fluorescent stem cell lines in order to examine the neurons as they mature during neuronal differentiation. High-quality images were acquired using confocal microscopy for multiple time points throughout development of AS, Dup15q, and control neurons. We found that by 12 weeks of age, AS neurons show a severe reduction in dendritic spines while Dup15q show an increase of spins compared to control. These deficits remain until at least 18 weeks in culture. Additionally, AS neurons have smaller somas than compared to control. These data provide a first glimpse of the development of human AS and Dup15q neurons.

Connecticut Regenerative Medicine Fund & National Institute of General Medical Sciences (P41 GM103313).

Symposium C: Talk No. 11

Comparing wearable sensors and video behavior coding in monitoring movement patterns in children with cerebral palsy.

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The *purpose* of this study is to compare two methods of monitoring (wearable sensors and video recording) movement patterns in children with moderate to severe cerebral palsy (CP). Technological advances have become critical to understanding how various pathologies affect the population and what improves the lives of the individuals affected. Wearable sensors (WS) and video behavior coding (VBC) present as relatively new techniques to monitor movement patterns and have provided promising results in monitoring movement patterns in healthy adults, typical developed children, and children with mild disabilities. Due to the performance and proven success of these methods, there has become a recent interest to understand what monitoring techniques are available to monitor children with moderate to severe neuromotor disabilities such as CP. Children with CP face many challenges, most notably significant deficits in posture and movement. Daily activities such as sitting and maintaining upright posture present as a challenge to individuals affected by CP. The severity of motor disability can be quantified by the Gross Motor Functional Classification System (GMFCS). This scale ranging, I-V correlates to how ambulatory and functional a child with CP is. GMFCS I is regarded as highly functional and ambulatory whereas GMFCS V correlates to least functional and requiring assistance. Although this project works with children with severe neuromotor disabilities, the findings of this study may translate to monitoring movement in patients with similar functional challenges. Eight children between the ages 1-13 years with GMFCS III-V were observed under two conditions: with and without trunk support to facilitate movement. These conditions allowed a clear change in behavior for each child, thus allowing comparison of the sensitivity of WS and VBC for quantifying these changes. In the no support session, the child only had pelvic stabilization when seated on a bench, whereas during support, the child was provided pelvic stabilization and external trunk support as determined by the Segmental Assessment of Trunk Control (SATCo). WS display general movement patterns as it corresponds to the speed of movement of the targeted region (trunk, arms) whereas VBC shows specific movement patterns such as both hands raising, or both hands down and does not account for the speed in which the region is moving. Upon comparison, a one-tailed Pearson's Product moment correlation highlighted a moderate correlation ($r=.452$, $p=.039$) between evaluating trunk movement from WS and VBC. This demonstrated that WS highlighted a change in movement similar to VBC. However, when evaluating the arms, there was an inverse correlation ($r=-.604$, $p=.007$) noted between the two monitoring devices. This inverse correlation is significant as it highlights the differences in outcomes of the measuring devices. It is imperative that clinicians and researchers understand what each technique is measuring to assure accurate interpretations. Jerky, rapid movement may be misinterpreted as improved function. If increased speed or rapid change in movement direction is desired than the WS may be better, but for children with moderate to severe disability slow, steady, functional movements may be the first signs of improvement.

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Poster Session 1 / No. 1

The Function of Synaptic Adhesion Proteins at Outer Hair Cells of the Cochlea

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Normal hearing requires both inner and outer hair cells of the cochlea. The outer hair cells are necessary for auditory sensitivity, but are known to be particularly susceptible to death as a consequence of intense noise exposure or aging. Death of these hair cells results in irreversible hearing loss. The outer hair cells are known to physically move back and forth in a process referred to as electromotility which increases cochlear sensitivity to sound waves leading to improved hearing sensitivity. However, elucidating the function of the neuronal circuit between the afferent synapse and the brain has been a stalwart task for years. Some proposed functions of outer hair cells could be in conveying information about damaging levels of sound, or facilitating communication between hair cells to affect their function in auditory sensitivity. One such protein found exclusively in outer hair cells is likely to be a key player. C1QL1 (complement component 1, q sub-component like 1) is a pre-synaptically released protein which promotes synapse maintenance. Utilizing a tool designed to knockout the outer hair cell afferent synapses in mice, we found that the conditional knockout mice of C1QL1 have lower auditory thresholds, implying that they can detect sounds at lower intensity levels than wildtype mice. This suggests that the function of the outer hair cell afferents is to serve as a negative regulator of hearing, making this the first ever evidence regarding outer hair cell afferent function. Moreover, conditional knockout mice startled at high intensity sound levels more than wildtype mice, demonstrating further their altered hearing abilities. Therefore, our data shows that the presence/absence of *C1ql1* affects outer hair cell response to acoustic exposure, and helps elucidate the potential role(s) of the outer hair cell afferent synapse, and its role in regulating electromotility and auditory thresholds.

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Poster Session 1 / No. 2

Observational learning of a foraging scenario in rats

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Using observation to learn about our environment is beneficial from an evolutionary perspective. It is safer and more efficient for an organism than learning by trial and error. While clearly important, observational learning studies in rodents show mixed results.
We present data from an observational learning paradigm in which rats observe and learn from conspecifics finding a food reward location in a T-shaped maze.
We contrast a reference and working memory version of the task. In the reference version the correct location is fixed and the observing rat receives exposure to the correct response over many days. In the working memory paradigm the goal location changes pseudorandomly daily, thus the observer must be attentive/observing at a specific time.

Poster Session 1 / No. 3

Independent and cooperative roles of the ERK1/2-MAPK and PI3K-AKT-mTOR pathways during developmental myelination and in adulthood

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Myelination is a multistep process involving the proliferation of oligodendrocyte progenitor cells (OPC), timely differentiation into oligodendrocytes (OLs), ensheathment of axons, initiation of myelin wrapping, and finally expansion of myelin sheath during the peak of myelination, which is maintained throughout adult life. Multiple studies have shown that the PI3K-Akt-mTOR and ERK1/2-MAPK signaling pathway play important, often overlapping, roles in the regulation of myelination. However, it is not well understood whether these two pathways independently regulate the distinct stages of oligodendrocyte differentiation and myelination or cooperate with each other at one or more levels of the signaling cascade. To address these questions we developed, and analyzed series of transgenic mice and asked whether the deficit in myelin growth caused by the loss of Erk1/2 signaling in oligodendrocytes could be restored to normal by hyper-activation of AKT in the Erk1/2 deficient OLs. Conversely, we investigated whether the inhibition of OPC differentiation and the hypomyelination caused by the loss of mTOR signaling could be restored to normal by hyper-activation of the Mek-Erk1/2 during the developmental myelination. We found that the hyperactivation of Akt in oligodendrocytes was able to partly rescue the deficits in myelin gene expression and myelin thickness in mice lacking ERK1/2. However, hyper-activation of ERK1/2 was unable to rescue the deficits in oligodendrocyte differentiation or hypomyelination caused by the lack of mTOR during early developmental myelination. We also examined whether mTOR signaling, as previously shown for Erk1/2, plays a role in myelin maintenance in the adult CNS. We found that the loss of mTOR activity by rapamycin treatment to adult mice led to minimal downregulation of myelin gene expression, compared to the sever deficit caused by the conditional ablation of Erk1/2 in oligodendrocytes of the adult mice. Together, our studies demonstrate that (1) PI3K-Akt-mTOR pathway, not the ERK1/2 MAPK pathway is independently regulates oligodendrocyte differentiation and initiation of myelination, (2) Both pathways cooperate to regulate myelin thickness during active myelination, (3) ERK1/2-MAPK signaling is mainly required for the long-term maintenance of myelin and axonal integrity with minimal contribution of mTOR in the adult CNS.

Poster Session 1 / No. 4

Molecular basis of antidromic rectification of gap junctions between AVA interneurons and motor neurons in *C. elegans* escape circuit

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C. elegans AVA command interneurons play a central role in escape behavior, and control A-type cholinergic motor neurons (A-MNs) through both electrical and chemical synapses. Our recent study shows that the gap junctions (GJs) between AVA and A-MNs conduct only antidromic currents (from A-MNs into AVA), and that the function of these GJs depends on UNC-7 innexin in AVA and UNC-9 innexin in A-MNs (Liu et al., *Nat Commun* 2017). However, molecular basis of the antidromic rectification is unknown. To answer this question, we began by expressing UNC-7 and UNC-9 in *Xenopus* oocytes, and analyzing biophysical properties of homotypic and heterotypic GJs formed by them. While UNC-9 has only one isoform, UNC-7 has ten different isoforms (UNC-7a through UNC-7j), which differ mainly in the length and amino acid sequence of the amino terminal. We found that various UNC-7 isoforms could independently form homotypic GJs with distinct properties in junctional voltage (V_j) dependence and junctional current (I_j) inactivation rate. In addition, all the UNC-7 isoforms could form rectifying heterotypic GJS with UNC-9 but the degree of rectification differed greatly. Several of these UNC-7 and UNC-9 heterotypic GJs essentially allowed a unidirectional current flow from the UNC-9 oocyte to the UNC-7 oocyte, which is like the direction of the I_j between AVA and A-MNs. Independent knockout of the specific UNC-7 isoforms conferring strong rectification caused a great inhibition of the I_j between AVA and A-MNs. Our results suggest that the GJs between AVA and A-MNs consist of UNC-9 in A-MNs and specific UNC-7 isoforms in AVA, and that the amino terminal domain of UNC-7 plays important roles in GJ gating.

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Poster Session 1 / No. 5

Establishing a molecular phenotype for Angelman Syndrome stem cell-derived neurons

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Angelman Syndrome (AS) is a neurodevelopmental disorder occurring approximately once in every 15,000 live births, characterized by severe seizures, absent speech, motor dysfunction, profound intellectual disability, and happy demeanor. Loss of expression of the maternal copy of *UBE3A*, a gene regulated by tissue-specific genomic imprinting, causes AS. *UBE3A* encodes an E3 ubiquitin ligase that may also act as a transcriptional co-activator. Although there is currently no cure, multiple therapies for AS, including gene therapy, are currently being explored. It will therefore be necessary to test the efficacy of these therapies in human AS neurons. Here, we propose to use isogenic AS and control stem cell-derived neurons to establish a molecular transcriptome phenotype for AS. We have generated two AS/control isogenic stem cell line pairs. First, we have derived induced pluripotent stem cells (iPSCs) from an AS patient with a missense mutation in *UBE3A* (F583S). This mutation does not affect UBE3A RNA or protein levels but causes a reduction in the protein's ubiquitin ligase activity, as demonstrated by *in vitro* ubiquitination assays. Using CRISPR/Cas9-mediated genome editing, we have corrected this point mutation in the AS iPSCs, generating an isogenic control iPSC line (F583S-CTRL). Second, we have used CRISPR/Cas9 to delete the maternal copy of UBE3A in H9 human embryonic stem cells (hESCs). Both isogenic pairs of stem cells have been successfully differentiated into forebrain neurons. Whole-cell patch-clamp recordings were performed on 12 week neurons to determine the extent to which the AS stem cell-derived neurons recapitulate the electrophysiological phenotypes seen in other non-isogenic AS iPSC-derived neuron lines. Stand-specific mRNAseq was performed on F583S and F583S-CTRL neurons and showed that 742 genes were differentially expressed in the F583S AS neurons (padj < 0.001). Lastly, we tested the ability to rescue electrical and molecular phenotypes by treating F583S and F583S-CTRL neural progenitors with lentivirus encoding E6 oncoprotein, which restores ubiquitin ligase function to the F583S mutant version of UBE3A in *in vitro* ubiquitination assays. Preliminary data suggests that restoration of ubiquitin ligase function following E6 treatment is sufficient to at least partially rescue AS phenotypes in iPSC-derived neurons. These results demonstrate that the transcriptome phenotype is a useful tool assaying phenotypic restoration in stem-cell derived neurons, and also suggest that activation of UBE3A may be a useful mechanism to explore for potential AS therapies.

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Poster Session 1 / No. 6

Cellular Senescence in iPS-derived Neural Progenitor Cells from Primary Progressive MS Patients Underlies Disease-related Defect in Myelination

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Primary progressive multiple sclerosis (PPMS) is a chronic progressive demyelinating disease of the central nervous system (CNS) without any effective treatment. To examine the potential for brain repair as a treatment strategy for PPMS we developed iPS cells from blood and skin samples of PPMS patients and age matched non-disease controls. Neural progenitor cells (NPCs) from controls protected against cuprizone, a myelin injury model, whereas PPMS NPCs failed to provide neuroprotection. We examined the effect of NPCs on oligodendrocyte progenitor cell (OPC) maturation and found that conditioned media from PPMS NPCs did not promote oligodendrocyte maturation *in vitro*. Analysis of PPMS NPCs identified elevated expression of hallmarks indicating cellular senescence, a cell stress response that results in an adaptive cellular physiology associated with aging, but also linked to degenerative disorders of the CNS. PPMS NPCs expressed elevated expression of $p16^{Ink4a}$, p53, HDAC2, senescence-associated β-galactosidase (SA-β-gal) activity, shortened telomeres, and production of factors related to a senescence-associated secretory phenotype (SASP). Using rapamycin, a known inhibitor of senescence, we found that the expression of these hallmarks could be reversed in the PPMS NPCs. Reversal of cellular senescence in PPMS NPCs reversed the deficit of oligodendrocyte differentiation *in vitro*. Proteomic analysis of the secretome of PPMS NPCs when compared with rapamycin treatment and age-matched non-disease control NPCs identified common and novel SASP factors, that are being investigated for potential roles in regulating OPC differentiation. Based on these data, we hypothesize that cellular senescence is responsible for the impaired function of PPMS NPCs on OPC maturation. These data for the first time reveal a mechanism by which NPCs via secretion of factors prevent endogenous remyelination in white matter lesions. While MS is diagnosed most often among young adults, at the cellular level, our data would suggest that MS may also be a disease of aging which may impact disease modeling and future treatment strategies.

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Poster Session 1 / No. 7

Firing characteristics of dorsal and ventral place cells in response to spatial novelty

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In both humans and rats, the hippocampus is important for memory and spatial navigation. Hippocampal neurons fire in specific physical locations generating a representation of the environment. However, if the environment or behavioral context is altered, a place cell can change its firing pattern. The behavioral correlates of place cell firing in dorsal hippocampus have been studied extensively, less is known about ventra cells. Firing characteristics will be determined while cells are recorded simultaneously from both regions as rats traverse a maze for a food reward.
(1) What are the basic firing characteristics of the place cells?
(2) To what degree is remapping correlated between regions?
(3) Do cells in each region respond differently to spatial novelty?

Poster Session 1 / No. 8

High sensitivity voltage-sensitive dyes for neuroscience research

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Voltage-sensitive dyes can be applied to brain cells and tissue to study the electrical signals used by the brain to function. The fluorescent dye molecules light up under a microscope and convert tiny electrical signals to measurable changes in light intensity. All neurological disorders involve changes in the brain's normal electrical activity and can potentially be studied with voltage-sensitive dyes. The Loew lab and their spin-off company Potentiometric Probes are working to develop voltage-sensitive dyes that are easy enough to use in virtually any academic or pharmaceutical industry research lab studying the brain's electrical signaling. To make this possible the dyes should be as sensitive as possible to voltage to provide a readout that is both very fast and clear. The Loew lab and Potentiometric Probes have developed a new voltage-sensitive dye technology that can potentially meet this goal and deliver the sensitivity required. If successful, this would make voltage imaging accessible to a much wider body of researchers and provide them with a more powerful tool for studying the electrical signals underlying normal and pathological brain function. One day, dyes may even be used in human patients to help diagnose and better treat neurological disorders.

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Poster Session 1 / No. 9

Glial Fibrillary Acidic Protein Rarely Colocalizes with Proliferation Markers in Adult Human Cortical Astrocytes

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Astrocyte intermediate filament Glial Fibrillary Acidic Protein (GFAP) expression is upregulated in response to disease and injury, and is commonly used as a marker for reactive astrocytosis. In order to determine whether GFAP is also a marker for proliferation in primary human cortical astrocytes, cells were placed in tissue culture, synchronized in G_0/G_1 and then followed for 24 and 48 hours after release back into the cell cycle. Immunocytochemistry was performed for GFAP and proliferation markers 5-bromodeoxyuridine (BrdU) and Ki67. 24 hours after release, 43.3% of astrocytes were proliferating, and 30.2% at 48 hours as observed by BrdU/Ki67 colocalization compared to total cells labeled with DAPI. BrdU labeling alone occurred in 11.9% of cells at 24 hours, and 25.7% at 48 hours, demonstrating cells returning to G_0 after mitosis. The total percentage of GFAP+ cells was 3.3% at 24 hours and reduced to 2.4% at 48 hours. Of the total cell population at 24 and 48 hours, ~1 in 725 proliferating cells colocalized with GFAP, while 76.8% of GFAP+ cells did not colocalize with proliferation markers at 24 hours, nor did 63.6% at 48 hours. Of the GFAP population that did colocalize with proliferation markers, 61.4% of cells colocalized with BrdU alone at 24 hours, and 41.3% with BrdU alone at 48 hours. These results indicate that 91.0% of cells labeling with GFAP at 24 and 78.6% at 48 hours were not in proliferation stages of the cell cycle. While human primary cortical astrocytes are entering the cell cycle at a high rate after release from G_0/G_1, GFAP+ cells are not representative of the proliferating population.

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Poster Session 1 / No. 10

Central neurons dictating locomotion are regulated by motor neurons through a feedback neural circuit

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The final output of a neural circuit depends on not only sequential synaptic transmission from higher to lower order neurons but also feedback regulation. It is well-established that feedback regulation can occur presynaptically through the activation of autoreceptors and transsynaptically through retrograde signaling. Studies of artificial recurrent neural circuits have led to the conjecture that feedback regulation from the final layer of output neurons to the upper layer of commanding neurons is crucial to circuit function in the nervous system of animal species. However, little, if any, experimental evidence exists to support this prediction. *C. elegans* locomotion neural circuit consists of a small number of premotor interneurons (command interneurons) and motor neurons. Synaptic transmission in this circuit occurs from premotor interneurons to cholinergic motor neurons, which, in turn, activate GABAergic motor neurons (D-MNs). A pair of premotor interneurons known as AVA play a central role in backward locomotion and escape response through activating a specific type of cholinergic MNs (A-MNs). Here we show that optogenetic activation of D-MNs strongly suppresses spontaneous excitatory postsynaptic currents (EPSCs) in A-MNs, and that either global knockout or AVA-specific knockdown of a specific $GABA_A$ receptor abolishes this inhibitory effect. In addition, we found that EPSCs in A-MNs are enhanced and locomotion waveforms are abnormal in worms with AVA-specific knockdown of the $GABA_A$ receptor. These observations suggest that motor neurons are not merely passive recipients of commands from higher order neurons but can profoundly regulate circuit functions through an inhibitory feedback circuit. This conclusion is supported by the worm connectome, which shows the existence of chemical synapses from D-MNs to the AVA premotor interneurons.

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Poster Session 1 / No. 11

FRS adapters show developmental stage specific selectivity in their ability to mediate FGFR signaling in the oligodendrocyte-lineage

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FGF signaling is important for numerous cellular processes and produces diverse cellular responses required in many tissues. Our recent studies using mice conditionally lacking *Fgfr1 or Fgfr2* during different stages of myelinogenesis reveled that FGFR signaling is first required embryonically for the induction of oligodendrocyte progenitors (Furusho et al., 2011) and then later postnatally for the growth of the myelin sheath during active myelination, but not for OPC proliferation, differentiation, or ensheathment of axons (Furusho et al., 2012). What intracellular signal transduction pathways recruited downstream of the FGFRs mediate these distinct developmentally regulated stage-specific responses remain unclear. Here we investigated the *in vivo* role of the adapter protein FRS2 (FGF-Receptor Substrate-2), considered a key immediate downstream target of FGF-receptors in the regulation of these responses at both the early and late stages of myelinogenesis. Using stage-specific mutants of *Frs2* (and *Frs3)* we found that they are together required for the specification of OPCs in the embryonic telencephalon at E12.5. Further, using an FRS2,3-binding site mutant of *Fgfr1*, we demonstrated that these FRS adapters are necessary for mediating all of FGFR1 signaling since the phenotype of these mutants mimicked that of the complete *Fgfr1* mutant. In contrast, during the postnatal period of active myelination, mutants with either conditional deletion of *Frs2/3* or with a point mutation of *Fgfr2* on the FRS2,3-binding site showed minimal deficits in myelin gene expression compared to that observed by loss of *Fgfr2*. This suggests that in mature oligodendrocytes FRS2/3 adapters are largely dispensable for transducing FGFR2-mediated signals, implying that at this stage of myelinogenesis other adapters are required for FGFR2 signaling. Together, our data suggests that the FRS adapters shows developmental stage specific ability to mediate FGFR signaling. Therefore, this contextual requirement for an intracellular transducer downstream of FGFR could in principle explain how FGF signaling plays distinct roles not only during different stages of myelinogenesis in the CNS but perhaps also in other developing systems.

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Poster Session 1 / No. 12

Anastellin: a TIMP-1 regulated fibronectin-derived peptide that inhibits oligodendrocyte maturation through regulation of S1P1 receptor function

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Astrocyte activation is associated with neuropathology and production of tissue inhibitor of metalloproteinase-1 (TIMP-1), an extracellular protein and protease inhibitor which we have shown to regulate oligodendrocyte progenitor cell (OPC) maturation and CNS myelination. To understand how TIMP-1 influences the astrocytic support for myelination, we interrogated the astrocyte secretome and performed comparative proteomic analyses of conditioned media (CM) from wildtype and TIMP-1 deficient (TIMP-1KO) primary astrocyte cultures. Tandem mass spectrometry (LC-MS/MS) of unique peaks identified by PF-2D proteomics identified a fibronectin (Fn)-derived peptide called Anastellin (Ana) in the TIMP-1KO secretome. Immunodepletion of Fn from TIMP-1KO astrocyte CM was found to restore OPC differentiation. To determine whether Ana could directly impact OPC differentiation, we applied recombinant Ana to primary OPCs in culture which resulted in a concentration-dependent reduction in Olig2+/MBP+ mature oligodendrocytes. This inhibitory effect of Ana that was further enhanced by coincident administration of Fn. Since Ana is known to act upon the spingosine-1-phosphate receptor 1 (S1PR1), we also determined that Ana attenuated the trophic effect of pFTY720 on OPC differentiation *in vitro*. Administration of FTY720 to either wildtype C57BL/6 mice during MOG-EAE induced clinical remission while FTY720 did not induce EAE disease attenuation in astrocyte-specific TIMP-1 deleted mice (GFAP-Cre-TIMP-1$^{fl/fl}$). Analysis of proteomic databases from human MS samples also identified Anastellin peptides in CSF of MS patients. Taken together, our data are consistent with a prominent role for astrocytic fibronectin in demyelinated lesions as an inhibitor of remyelination in MS. These results signify astrocytic production of TIMP-1 during neuroinflammation as a beneficial response to injury which impacts the lesion microenvironment in MS.

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Poster Session 2 / No. 13

Using Brainbow multispectral labelling to image disinhibitory cortical networks

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Information processing in the cortex involves modulation of excitatory outputs by inhibitory GABAergic interneurons. GABAergic interneurons encompass a diverse subset of cell types, with each having a distinct modulatory effect on its target cell. Unlike most GABAergic cells which attenuate excitation, vasoactive intestinal peptide (VIP) expressing GABAergic interneurons play a unique disinhibitory role in cortical circuitry and can therefore act to potentiate excitation. This disinhibition is due to VIP interneurons preferentially connecting onto somatostatin (SST) expressing GABAergic interneurons and decreasing the inhibitory effect of SST onto excitatory principal cells. While it has been established that VIP interneurons synapse onto SST cells, the specific convergence patterns of VIP cells onto their targets has not been worked out. Our approach is to use Brainbow to address the convergence of VIP interneurons onto regions of interest and eventually specific target cells of interest. Brainbow 3.0 makes use of a cre-driven adenoassociated virus and through additional immunostaining, we will determine the number and location of unique VIP synapses that occur. This method results in intense multicolored labelling, which allows us to determine presynaptic density and the number of different cells synapsing within a given area. Preliminary results indicate that there are upwards of 150 disinhibitory presynaptic terminals within a 57.5mm volume of cortical tissue. We demonstrate that Brainbow can be used quantitatively and that measurement of the density of presynaptic terminals can be informative towards resolving details underlying cortical disinhibition.

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Poster Session 2 / No. 14

Psychophysiological Response to a Discarding Task in Hoarding Disorder

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Hoarding Disorder (HD) is characterized by an inability to discard items leading to excessive clutter. Little research has been done in regards to the autonomic and psychophysiological response in HD, especially when making decisions on whether to discard or keep an item. Research has shown that discarding personal items is associated with abnormal activation in the brain regions of decision-making as well as self-reported distress, compared to that of a control item. In this study we aimed to compare psychophysiological response when discarding personal vs. control items. Additionally, we compared psychophysiological responses during discarding between hoarding patients who had previously completed cognitive-behavioral treatment and those who were treatment naïve. Patients with a primary diagnosis of hoarding disorder completed self-report measures of HD symptoms and then completed a discarding task, which consisted of sorting and discarding their own possessions as well as control items (the "experimenter's" mail) End tidal carbon dioxide ($ETCO_2$), heart rate (HR) and heart rate variability (HRV), and skin conductance level (SCL) data were collected throughout the task. Preliminary results (current n=38, projected N=53) showed that discarding personal mail resulted in lower $ETCO_2$, $t(35) = -5.02$, $p < .001$, $d = -0.82$, greater HR, $t(34) = 2.49$, $p = .018$, $d = 0.36$, and greater SCL, $t(33) = 3.34$, $p = .002$, $d = 0.64$ compared to discarding the experimenter's mail. HRV was not different between mail types, $t(34) = 0.42$, $p = .676$. No significance was found in patients who had previous cognitive-behavioral treatment compared to the treatment naïve. Similarly, psychophysiological responses during discarding tasks were not correlated with the self-reported HD symptoms. The results suggest that discarding personal possessions leads to greater psychophysiological activation compared to discarding of others' possessions. Should the results be continually supported in the full sample, it will allow for greater understanding of the psychophysiological mechanisms of HD, and lead to better understanding of how to treat patients with this disease.

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Poster Session 2 / No. 15

Role of miR-181c-5p in post-stroke social isolation

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Dysregulation of microRNAs (miRNAs) have been tied to several neurological disorders including stroke. MiRNA profiles can also change as a result of social environments. We have previously shown that the social isolation is detrimental to post-stroke recovery. miR-181c-5p emerged as one of the few miRNAs modulated by both stroke and social environment. Therefore, in this study we examined the potential role of miR-181c-5p after post-stroke social isolation (SI).Dysregulation of microRNAs (miRNAs) have been tied to several neurological disorders including stroke. MiRNA profiles can also change as a result of social environments. We have previously shown that the social isolation is detrimental to post-stroke recovery. miR-181c-5p emerged as one of the few miRNAs modulated by both stroke and social environment. Therefore, in this study we examined the potential role of miR-181c-5p after post-stroke social isolation (SI).

2-3 month-old C57BL/6 male mice were pair-housed (PH) for at least two weeks. After two weeks, a 60-minute stroke was induced and mice were randomly assigned to one of two housing conditions, isolation or continued pair-housing (ST-ISO or ST-PH). In the treatment study group, ST-ISO mice were randomized to receive either miR-181c-5p mimic (7mg/kg i.v/day x drug) or a vehicle control administered through lateral tail vein at 24h and 48h after stroke. The effect of mimic treatment was evaluated at 1, 3, and 7 days after stroke using rotarod, open field test (OFT) and corner test. Mice were sacrificed 7 days after stroke for miRNA analysis. Target genes for miR-181c-5p were analyzed by qPCR using an RT2 Profiler qPCR Array of pre-coated miR-181c gene target by Qiagen.

Temporal profile expression data suggests that miR-181c-5p was significantly down regulated ($p<0.05$ vs ST-PH) up to 7 days after post-stroke social isolation. Treatment with mimic significantly increased miR-181c-5p expression in the brain tissue. Further social isolation-induced reduction in miR-181c-5p expression was rescued by mimic treatment. Post treatment with mimic partially restored sensori-motor deficit in post stroke socially isolated mice. Target gene analysis identified several genes related to calcium signaling e.g. *Cpne2* and *Enkur* that were downregulated after mimic treatment.

In summary, our temporal profile expression data suggests that miR-181c can be potential target for the treatment of post stroke SI. Treatments with miR-181c mimic upregulated miR-181c- 5p expression in the brain tissue suggesting its efficacy in crossing blood brain barrier. Mimic treatment also partially restored sensori-motor deficits in post stroke SI mice. Our preliminary data suggest that genes related to calcium signaling might be involved in the beneficial effect of miR- 181c mimic, although additional confirmatory studies are needed.

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Poster Session 2 / No. 16

Using Peptides to Dissect the Role of Kal7/PSD95/NR2B Interactions in Synaptic Plasticity in CA1 Pyramidal Neurons (475 words)

Mason L. Yeh, Eric S. Levine, Elizabeth A. Eipper, Richard E. Mains

The Kalirin (KALRN) gene has been implicated in schizophrenia, autism, stroke, substance abuse and intellectual disability. Alternative splicing results in three major isoforms - Kal9, Kal12 and Kal7, with expression of Kal7 predominant in the mature central nervous system (CNS). Kal7 is concentrated at the postsynaptic density (PSD). Evoked field potential recordings in Kal7 knockout mice, which lack only this isoform, revealed its essential role in NMDA receptor-dependent forms of long-term potentiation (LTP) and long-term depression (LTD) in the hippocampus. The expression of LTP and LTD in hippocampal neurons forms the cellular basis of learning and memory. Therefore, identifying the targets and signaling cascades affected by the absence of Kal7 may provide a better understanding of the pathophysiology of several neurological disorders. Biochemical analyses identified specific regions of Kal7 that interact with PSD95, a major PSD scaffolding protein, and with the GluN2B subunit of NMDA receptors. Using peptides designed to disrupt only the Kal7/PSD95 interaction, the NR2B/PSD95 interaction or the Kal7/NR2B interaction, we set out to determine whether acute disruption of specific aspects of Kal7 signaling in otherwise wild type neurons would affect the expression of synaptic plasticity (LTP and/or LTD) in hippocampal CA1 neurons of young adult mice. This unique approach may have broad implications in disease states which may affect Kalirin signaling in the mature CNS, as opposed to knockout mouse models, which lack Kal7 in all neurons since conception. We employed whole-cell patch clamp electrophysiology and biochemical assays to assess synthetic peptides identical to (1) the PDZ-binding motif of Kal7, (2) the PDZ-binding motif of GluN2B and (3) the region of the Kal7 domain which binds to a juxta-membrane region of GluN2B. For electrophysiological recordings, peptides were individually added to the intracellular recording solution at a concentration of 1-10 μM and allowed to fill a postsynaptic neuron situated in CA1 prior to administering theta-burst stimulation (TBS) to the Schaffer Collaterals. Single-evoked excitatory postsynaptic currents (eEPSCs) were recorded for 10 minutes prior to TBS and for 60 minutes post-TBS. All 3 peptides blocked the induction of LTP in a dose-dependent manner; a mutant Kal7 PDZ-binding motif peptide lacked this effect. The ability of high-frequency stimulation (HFS, tetanus), 100 Hz for 4 seconds, to elicit LTP in CA1 pyramidal neurons was still observed in neurons filled with peptides mimicking the PDZ-binding motif of Kal7 or the PDZ-binding motif of GluN2B. In addition to their interaction with each other, both Kal7 and GluN2B interact with the PDZ domains of PSD95. Using biotinylated peptides we set out to determine the binding affinities of Kal7 and GluN2B with the three PDZ domains of PSD95 (PDZ1, PDZ2, PDZ3 and PDZ123). Collectively, we present a novel method of interrogating Kal7 and GluN2B interactions with PSD95 in the PSD of pyramidal neurons located in CA1 of the hippocampus, and their roles in the induction and maintenance of synaptic plasticity.

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Poster Session 2 / No. 17

BDNF causes release of endogenous cannabinoids at CA1 inhibitory synapses

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Endocannabinoids (eCBs) are retrograde messengers that act on presynaptic terminals to suppress neurotransmitter release. Brain derived neurotrophic factor (BDNF), traditionally known for promoting neuronal growth and development, is also a modulator of synaptic transmission. Both BDNF and eCBs are involved in higher cognitive functions. The effects of eCBs and BDNF are mediated by cannabinoid receptor type 1 (CB1R) and trkB receptors respectively. Both the receptors are highly expressed in cortical layers II/III and V. Our lab has previously shown that, at inhibitory synapses of layers II/III of somatosensory cortex, BDNF induces post-synaptic release of eCBs that act retrogradely at presynaptic terminals to suppress GABA release. Further, we have shown that BDNF triggers the production of 2-AG via BDNF/trkB/PLC signaling to exert its effects on GABAergic transmission. We know that (1) CB1 and trkB receptors are abundantly expressed in hippocampus and (2) both eCBs and BDNF regulate neurotransmission in hippocampus. In the current study, we explored a potential interaction between BDNF and eCBs at CA1 inhibitory synapses of hippocampus. We hypothesize that, similar to somatosensory cortex, BDNF will induce postsynaptic release of eCBs that act retrogradely to suppress GABA release at inhibitory synapses. We found that acute application of BDNF reduced spontaneous inhibitory postsynaptic currents (sIPSCs). This effect of BDNF was completely blocked by a trkB receptor antagonist suggesting that the effect is triggered by trkB activation. The suppressive effects of BDNF also require eCB signaling, as the effect of BDNF on sIPSCs was prevented by a CB1 receptor antagonist. Further, we disrupted the effect of BDNF on sIPSCs by blocking eCB transport and re-uptake. Blocking eCB transport prevented the effect of BDNF while re-uptake inhibition enhanced the effect of BDNF. These results suggest that BDNF triggers post-synaptic release of eCBs. To identify the specific eCB release by BDNF, we tested the effects of disrupting the synthesis and degradation of the eCBs, 2-AG and anandamide. A DAG lipase inhibitor, which blocks the synthesis of 2-AG, completely blocked the effect of BDNF. In addition, an inhibitor of the 2-AG degrading lipase, monoacyl glycerol lipase (MAGL), which would increase 2-AG levels, enhanced the effect of BDNF. In contrast, there was no change in BDNF's effect when we blocked anandamide degradation with an inhibitor of fatty acid amide hydrolase (FAAH). Collectively, these results suggest that in the hippocampus, BDNF-trkB signaling induces the release of the endogenous cannabinoid 2-AG, which acts as a retrograde messenger at presynaptic CB1 receptors to suppress GABA release. Future studies will address the interaction of BDNF and eCBs at CA1 excitatory synapses.

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Poster Session 2 / No. 18

Applying rule-based modeling to understand UBE3A's role in dendritic spine morphogenesis.

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UBE3A encodes for the ubiquitin ligase UBE3A which targets substrates to be degraded by the 26S proteasome. In neurons, only the maternal copy of *UBE3A* is expressed due to genomic imprinting. The loss of the maternal copy of *UBE3A* results in Angelman Syndrome (AS), a neurodevelopmental disorder characterized by absent speech, motor dysfunction, severe seizures, intellectual disability, and a happy demeanor. Despite the cause of AS being well established, the mechanism behind UBE3A is still unknown. Multiple targets of UBE3A have been proposed, but many have failed to be found in neurons. The lack of ubiquitination and consequently degradation of UBE3A substrates is predicted to increase the substrate protein levels in AS. We have developed a rule-based model to investigate UBE3A's role in protein degradation and possible relation to dendritic spine morphogenesis. Our model encompasses the ubiquitin proteasome pathway (UPP), co-activators of UBE3A, possible UBE3A substrates, and upstream signaling of the UPP. We have modeled an AS patient's point mutation in UBE3A's HECT domain which diminishes ubiquitin ligase activity. *In vitro* assays have shown that the ubiquitin ligase activity of this mutant can be rescued with E6, a co-activator of UBE3A. However, our model does not show the same robust results in the presence of the entire UPP. When we simplify our model to only have the same molecules as the *in vitro* assays we successfully recapitulate the results. Our model mimics the current issue in the field which is confirming UBE3A substrates *in vivo.* Our goal is to use model to understand the differences seen between *in vivo* and *in vitro* experiments as well as tease apart how co-activators could restore ligase activity.

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Poster Session 2 / No. 19

Light-induced activation of dopamine and GABA release on striatal medium spiny neurons in brain slices

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The nigrostriatal pathway, originating in the substantia nigra (SN) and terminating in the striatum, plays a critical role on cognitive and motor functions relevant to a variety of disorders including Parkinson's disease, addiction, and schizophrenia. Striatal medium spiny neurons (MSNs) bear dopaminergic receptors whose activation influences behavior and cognition. A transgenic mouse with light-sensitive channels in dopaminergic neurons (DAT-ChR2) is routinely used to evaluate the physiology of dopamine (DA) neurotransmission. The present study explores the simultaneous transmission of DA and GABA, upon blue light (475 nm) activation of DAergic axons in the striatum (brain slices) by whole-cell recordings of the postsynaptic responses in MSNs. Repetitive light activation of striatal DAergic fibers revealed a strong depression of the GABA output from these fibers, with a relative refractory period of approximately 10 s. However, successive light pulses on the cell bodies of DAergic neurons in SN showed a 100-fold shorter refractory period (100 ms) for firing somatic action potentials. These differences in recovery time suggest inefficient transmission of information from DA axons to MSNs, or likely the result of severing the axons in slice preparations. A pharmacological block of DA receptors did not produce any effects on light-induced synaptic currents in MSNs. Despite both GABA and DA being stimulated by the same pulse of light, DA release appeared not to have any effect on the postsynaptic GABA currents. Further exploration of light- and synaptically-induced DA-GABA co-release will allow for a more thorough understanding of the complex relation between GABA and DA on neuropsychiatric disorders influenced by the nigrostriatal pathway.

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Poster Session 2 / No. 20

Schwann cells: De-differentiation into Myofibroblasts during Injury/Fibrosis

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The transparent cornea is the most innervated tissue in the human body. Corneal transparency is maintained by a precise spatial distribution of its cellular components, including the resident stromal cells (keratocytes), and the non-myelinating corneal Schwann cells (nmcSCs), glial cells devoted to the maintenance, and health of the stromal nerves. Surgical procedures involving deep corneal stromal incisions can lead to axonal degeneration, and corneal opacification. Upon insult(s), keratocytes differentiate into myofibroblasts, a contractile mesenchymal cell type. Myofibroblasts are needed for repair; however, their persistence in the corneal tissue can lead to corneal opacity, and fibrosis. Myofibroblasts can also derive from bone marrow precursor cells. Our lab made the discovery that a new source of myofibroblasts is involved in the post-injury process: the nmcSCs. Now, using a mouse model of deep stromal injury, we propose to characterize the cellular process of trans-differentiation from nmcSCs to myofibroblasts, and their contribution to corneal opacity. Corneas will be collected at different time points post-injury, and samples will be processed for immunohistochemistry analysis for markers of fibrosis including □-smooth muscle actin (□SMA), and vimentin (Vim), and marker of nmcSCs (Glial Fibrillary Acidic Protein, GFAP). We hypothesize that, after injury, $GFAP^+$ cells will also express □-SMA and Vim; as the cornea heals, the number of $GFAP^+$/ □SMA^+/Vim^+ cells will be reduced. However, if the repair process is not complete, $GFAP^+$/ □SMA^+/Vim^+ cells will be still present in the cornea, and cause opacity. This study illuminates, for the first time, a potential role of nmcSCs in either corneal repair or pathologies.

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Poster Session 2 / No. 21

Expression of Connexin Genes in the Human Developing Cortex at Mid-Gestation

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In the developing human brain, neuronal physiological activity does not depend entirely on sensory inputs. Our group has previously demonstrated that connexin hemichannels contribute strongly to spontaneous depolarizations in the human fetal cortex at mid-gestation (17 to 23 gestation weeks). In order to understand the role of specific connexins in the developing brain, it is important to decipher their spatial and temporal pattern of expression. In the present study, we used tissue samples from postmortem human fetal cortex at mid-gestation to determine the expression of connexin isoforms Cx26, Cx36 and Cx45 at the transcriptional level with RNA sequencing and, at the translational level by immunolabeling. We found that the level of gene expression and protein expression of Cx45 was the highest, while that of Cx26 was the lowest among the three “neuronal” connexin isoforms (Cx26, Cx36 and Cx45). It is important to emphasize that the supremacy of Cx45 was determined independently by RNA sequencing and by immunolabeling. Our RNA sequencing results enabled us to assess the expression of additional connexin isoforms (Cx30, Cx31.9, Cx32, Cx40, Cx40.1, Cx47), as well as two pannexin isoforms (PANX1 and PANX2). We found that Cx43, Cx36, PANX1 and PANX2 were in the range between 2 - 10 rlog units, while Cx32, Cx40.1 and Cx31.9 were approximately 25-fold lower, in the range 0.1 - 0.4 rlog units. Our RNA sequencing data are thus consistent with enhanced neurogenesis during the mid-gestation stage of the human fetal brain development, as observed by increased expression of pannexin-1 (PANX1) along with “neuronal” connexin isoforms Cx45 and Cx36. Moderate Cx47 and Cx43 expression levels suggest that postmitotic oligodendrocytes and postmitotic astrocytes may also be present at this stage of human development. Neuronal connexin subtype, Cx45, is the dominant isoform among all connexin proteins in the human fetal cortex during mid-gestation, 17th to 23rd gestational week.

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Poster Session 2 / No. 22

Spontaneous Calcium Signals and Electrical Activity of Cortical Human Neurons During *In Vitro* Differentiation

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Spontaneous calcium signals are an important subset of factors that drive developmental processes, including proliferation, migration, and neuronal differentiation. Therefore, the cellular and molecular mechanisms underlying calcium influx through plasma membrane, calcium release from intracellular stores and other forms of intracellular calcium signaling, have a very important role during development of functional neurons. On the other hand, one of the main characteristics that distinguishes neurons from other cell types in the brain is their ability to fire action potentials, the property dependent on the presence of voltage-gated sodium and potassium channels. The expression of voltage-gated channels is gradual during the early stages of neuronal differentiation. Once established, this feature is considered to be of great importance during the remaining phases of neuronal and brain functional development (e.g. axon growth, dendrite and spine sprouting, synaptogenesis, network refinement, etc.). The aim of this study is functional characterization of electrical properties and calcium signaling during early *in vitro* differentiation of cortical neurons from human fetal neural precursors. Electrical properties of cells were evaluated by whole-cell patch clamp technique, while the detection of spontaneous calcium signals was done by multisite optical imaging using a membrane permeable calcium-sensitive dye Oregon Green Bapta-1 AM, LED illumination (470 nm) and RedShirtImaging CCD camera. The source of ionic calcium underlying spontaneous activity, as well as the responsible plasma membrane channels/receptors involved in both electrical activity and calcium transients, were analyzed by the application of pharmacological agents that block voltage gated sodium channels, calcium channels, intracellular calcium stores, transient receptor potential channels, gap junctions, and connexin hemichannels.

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Poster Session 2 / No. 23

Biochemical Analysis of the Synaptic Regulator C1QL3 and its Binding Partners

Matthew Sticco, Brianna Thompson, Susanne Ressl, and David Martinelli

Synapses are specialized neuronal cell-cell junctions, which allow for the formation of neural networks by promoting fast, efficient, and stable communication between neurons. C1QL3 is a secreted protein that is expressed in small subset of brain regions, mostly in the limbic system the adult brain. C1QL3 is a known high affinity ligand of BAI3, an adhesion-class GPCR that is located post-synaptically when expressed in the central nervous system. It has been shown that C1QL3 regulates synapse formation and/or maintenance by conditional C1QL3 knockout mice. This knockout also results in abnormalities in fear memory likely due to the reduction in synapse number after C1QL3 knockout. The mechanism by which C1QL3 influences synapse density is currently unknown but could involve a trans-synaptic adhesion complex between C1QL3, BAI3, and an identified, but yet-to-be-validated, presynaptic binding partner, NPTX1. Another protein, Spock2, is thought to be a negative regulator of this complex. Through cell surface binding with a series of site-directed mutagenesis experiments, we hope to reveal the binding surfaces of C1QL3 and validate the identity of its pre-synaptic binding partner. In addition, we hope to determine binding constants for each C1QL3 binding partner. We also aim to show this complex in vivo using three-color immunofluorescence combined with expansion microscopy to enhance optical resolution. This may provide insights about how C1QL3 influences the formation and/or maintenance of synapses.

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Poster Session 2 / No. 24

Pharmacological and genetic studies of effort-related decision making using mouse touchscreen procedures: Effects of dopamine antagonism and humanized catechol-o-methyltransferase variants.

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Effort-based decision making tasks offer animals choices between a more preferred reinforcer that requires high effort to obtain vs. a low effort/low reward option. The neural mechanisms of effort-based choice have been widely studied in rats, and evidence indicates that a distributed forebrain circuit that includes mesolimbic dopamine (DA) and related neural systems plays a key role. DA antagonism or depletion produces a low-effort bias in rats tested on effort-based choice tasks. However, fewer studies of effort-based choice have been performed in mice, and most of these have involved T-maze choice tasks. The present studies used touchscreen operant procedures (i.e., Bussey-Saksida boxes) to assess effort-based choice in mice. The high-effort component involved rearing up to press an elevated lit panel on the touchscreen in order to receive Ensure strawberry milkshake as the reinforcer. The low effort choice was a dish of less preferred food pellets (Bio-serv) that was freely available in the box. CD1 mice were tested in a series of experiments in which the mice pressed the panel on a fixed ratio 1 (FR1) schedule. Injections of the DA antagonist haloperidol (0.05-0.15 mg/kg IP) produced a dose-related decrease in panel pressing. Intake of the concurrently available food pellets was not reduced by haloperidol, and in fact, there was a significant quadratic trend, showing that there was a tendency for pellet intake to increase at the low/moderate doses. In contrast, reinforcer devaluation by prefeeding substantially decreased both panel pressing and pellet intake. In free-feeding choice tests, mice strongly preferred the Ensure vs. the pellets, and haloperidol had no effect on food intake or preference. Additionally, humanized catechol-O-methyl transferase (COMT) transgenic mice with two genotypes (Val and Met variants) as well as wild-type (WT) mice from the S129 background were tested using touchscreen choice procedures. Mice were trained in a FR/choice task with FR requirements varied (FR1, 2, 4, 8, to 16) in an ascending and descending sequence. Results show an inverse relationship between the number of reinforcers delivered by panel pressing and pellet intake across the different FR levels in all three groups. There was a significant group x FR level interaction, with panel presses in the Val group being significantly lower than WT group on FR1, 2, 4, and 8, and lower than Met group on FR4. These studies show that haloperidol did not reduce panel pressing due to decreases in primary food motivation or preference, and illustrate the possible relation between COMT polymorphisms and negative symptoms such as motivational dysfunctions in human psychopathologies.

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Thank you to all for attending this year's retreat!

Special thank you to all trainees, judges and vendors for their time and contributions to this event.

We hope you enjoyed yourselves!

LOOK FORWARD TO SEEING YOU NEXT YEAR!

Spring 2019