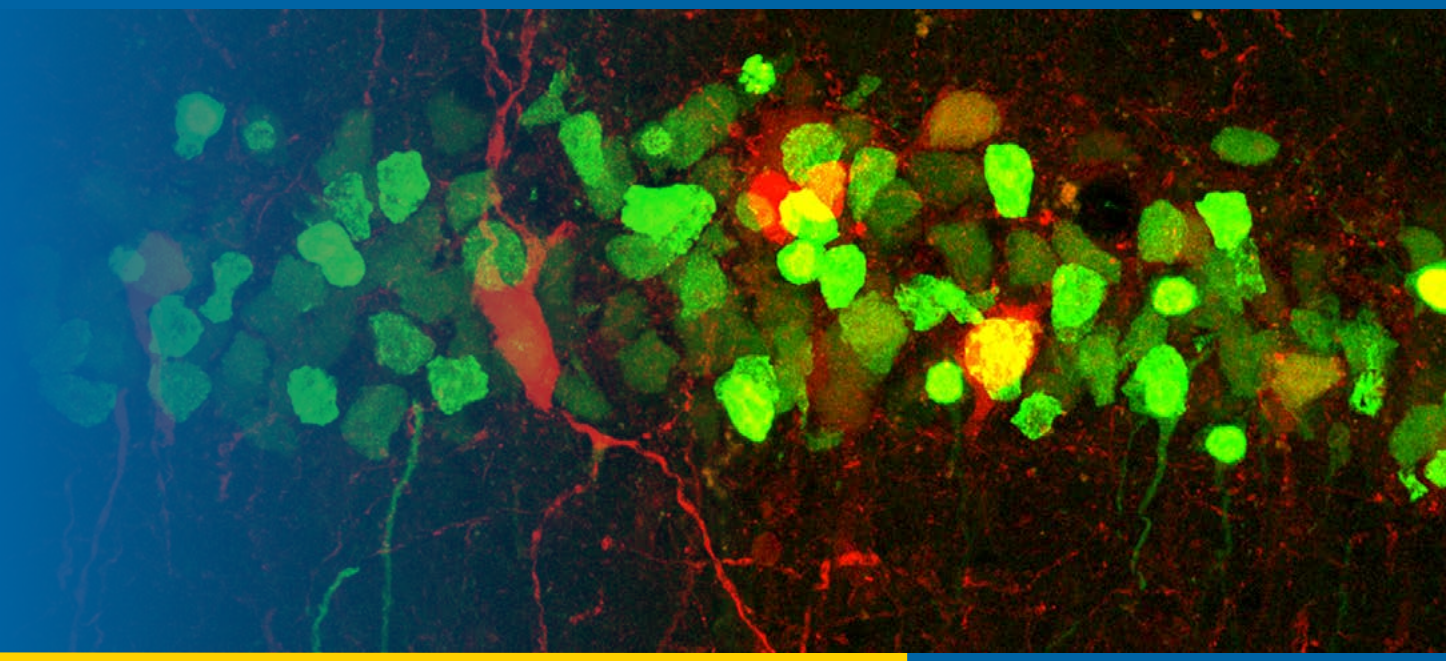
UCI Center for Neural Circuit Mapping | Cajal Club

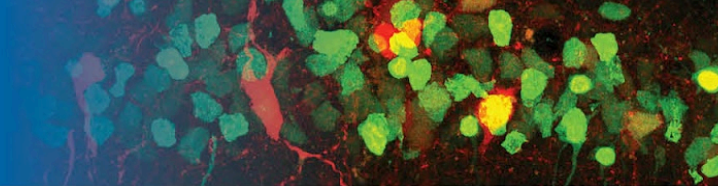


2024 CONFERENCE

Brain Cell Types, Circuits, and Disorders

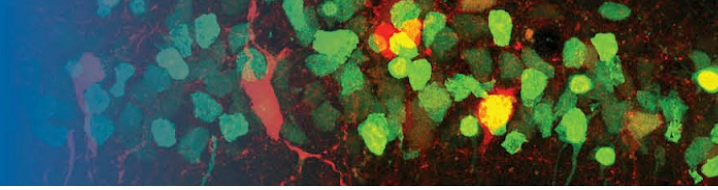
August 19 – 21
7:30 a.m. - 5:00 p.m.

Beckman Center of the National Academies of Science & Engineering, Irvine CA



2024 Conference Schedule

Day 1 - Monday, August 19	
7:30 – 8:15 a.m.	Breakfast and Registration
8:15 – 8:30 a.m.	Conference Introduction (Dean Michael J. Stamos)
Session 1: Brain Cell Types, Connectivity and Genomics	
8:30 – 8:35 a.m.	Session Introduction: Bing Ren, PhD
8:35 – 9:10 a.m.	Joe Ecker, PhD (Salk Institute)
9:10 – 9:45 a.m.	Bing Ren, PhD (University of California, San Diego)
9:45 – 10:20 a.m.	Mariano Gabitto, PhD (Allen Institute)
10:20 – 10:40 a.m.	Break
10:40 – 11:15 a.m.	Giorgio Ascoli, PhD (George Mason University)
11:15 – 11:50 a.m.	Elly Nedivi, PhD (Massachusetts Institute of Technology)
12:00 – 1:00 p.m.	Lunch and Poster Session
1:00 – 1:30 p.m.	Poster Session Continued
Session 2: Precision Brain Cell Access and Imaging	
1:30 – 1:35 p.m.	Session Introduction: Gordon Fishell, PhD
1:35 – 1:55 p.m.	TBD Short Talk
1:55 – 2:30 p.m.	Gordon Fishell, PhD (Harvard University)
2:30 – 3:05 p.m.	Tanya Daigle, PhD (Allen Institute)
3:05 – 3:25 p.m.	Break
3:25 – 4:00 p.m.	Scott Sternson, PhD (University of California, San Diego)
4:00 – 4:35 p.m.	Lin Tian, PhD (University of California, Davis)
4:35 – 5:10 p.m.	Mikhail G Shapiro, PhD (California Institute of Technology)
5:15 – 6:45 p.m.	On-site reception for all attendees



Day 2 - Tuesday, August 20

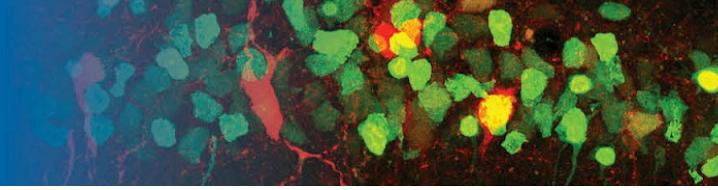
7:30 – 8:30 a.m.	Breakfast

Session 3: Neural Circuits and Networks

8:30 – 8:35 a.m.	Session Introduction: Larry Swanson, PhD
8:35 – 9:10 a.m.	Larry Swanson, PhD (University of Southern California)
9:10 – 9:45 a.m.	Hong-Wei Dong, PhD (University of California, Los Angeles)
9:45 – 10:20 a.m.	Li Zhang, PhD (University of Southern California)
10:20 – 10:40 a.m.	Break
10:40 – 11:15 a.m.	Wei Wei, PhD (University of Chicago)
11:15 – 11:50 a.m.	Hysell Oviedo, PhD (Washington University)
12:00 – 1:00 p.m.	Lunch and Poster Session
1:00 – 1:30 p.m.	Poster Session Continued

Session 4: Brain Disorders

1:30 – 1:35 p.m.	Session Introduction: Xiaoke Chen, PhD
1:35 – 1:55 p.m.	TBD Short Talk
1:55 – 2:30 p.m.	Xiaoke Chen, PhD (Stanford University)
2:30 – 3:05 p.m.	X. William Yang, PhD (University of California, Los Angeles)
3:05 – 3:25 p.m.	Break
3:25 – 4:00 p.m.	Nicholas Seyfried, PhD (Emory University)
4:00 – 4:35 p.m.	Monica Carson, PhD (University of California, Riverside)
4:35 – 5:10 p.m.	Kalpna Gupta, PhD (University of California, Irvine)
5:45 – 7:30 p.m.	Hosted Speaker Dinner located at Il Fornaio



Day 3 - Wednesday, August 21

7:30 – 8:30 a.m.	Breakfast

Session 5: Frontiers, New Concepts and Approaches

8:30 - 8:35 a.m.	Session Introduction: Jianhua Cang, PhD
8:35 – 9:10 a.m.	Jianhua Cang, PhD (University of Virginia)
9:10 – 9:45 a.m.	Joshua Trachtenberg, PhD (University of California, Los Angeles)
9:45 – 10:20 a.m.	Gianluca Tosini, PhD (Morehouse University)
10:20 – 10:40 a.m.	Break
10:40 – 11:15 a.m.	Xiaoning Bi, PhD (Western University)
11:15– 11:50 a.m.	Michael Koob, PhD (University of Minnesota)
12:00 – 1:00 p.m.	Lunch and Poster Session
1:00 – 1:30 p.m.	Poster Session Continued

Session 6: TBD and Viral Core Workshops

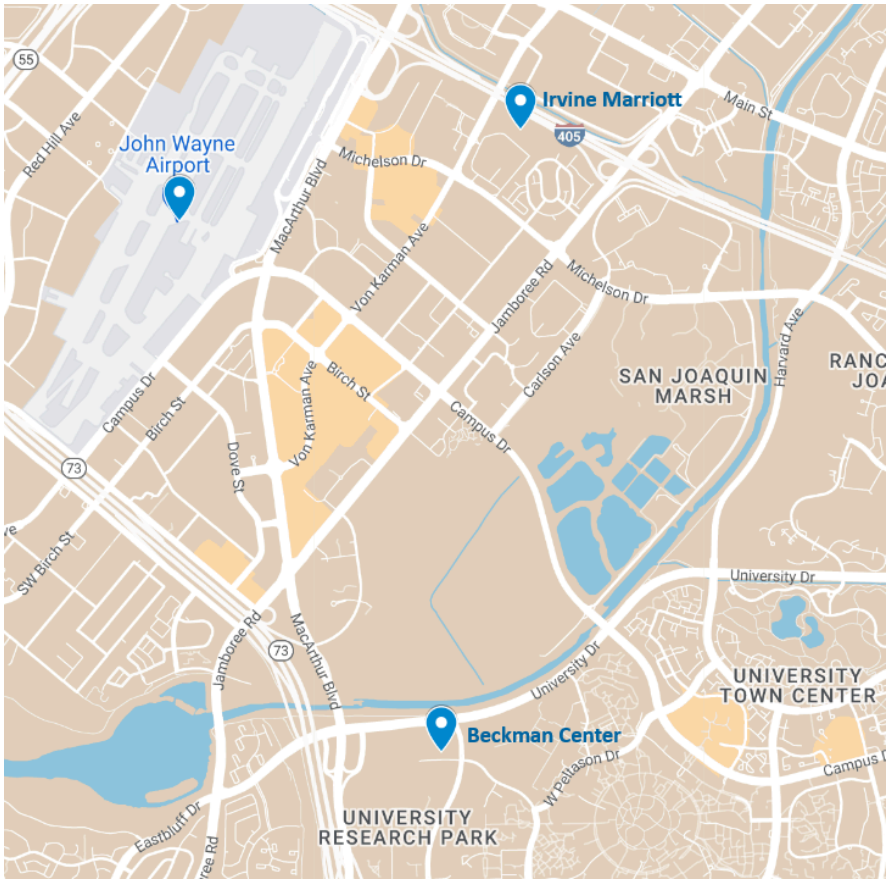
1:40 – 3:00 p.m.	Workshop presented by TBD
3:00 – 3:20 p.m.	Break
3:20 – 4:50 p.m.	Viral Vector lecture presented by the CNCM, Tim Shay, PhD (California Institute of Technology) Alexis Bouin, PhD (University of California, Irvine)
4:50 – 5:00 p.m.	Closing Remarks

We are pleased to hold our conference at the Arnold and Mabel Beckman Center of the National Academies of Sciences and Engineering. As a scientist and a businessman, Dr. Beckman envisioned a West Coast center where experts could discuss matters of science and technology. A grant from the Arnold and Mabel Beckman Foundation, and The Irvine Company's land donation, Dr. Beckman was able to make this vision a reality. The Arnold and Mabel Beckman Center of the National Academies of Sciences and Engineering opened its doors in April, 1988, and the Beckman Center continues to be operated by the academies as their West Coast location for both program activity and conferences.

Directions
100 Academy Way, Irvine CA.

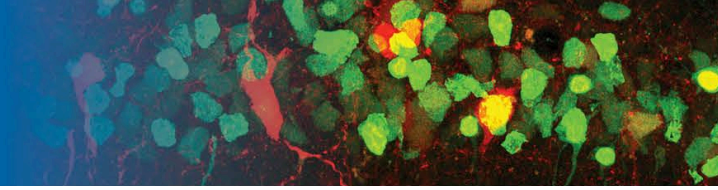
Near University Drive, exit from 73.

Adjacent to the University of California, Irvine and less than 3 miles from John Wayne Orange County Airport.

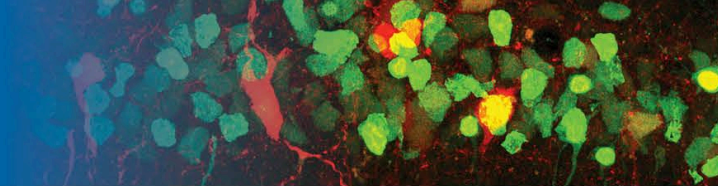


Parking

Parking is available on-site at no charge. Electric charging stations are available on-site. No overnight parking.



Session 1
Brain Cell Types, Connectivity and Genomics



Joe Ecker, PhD

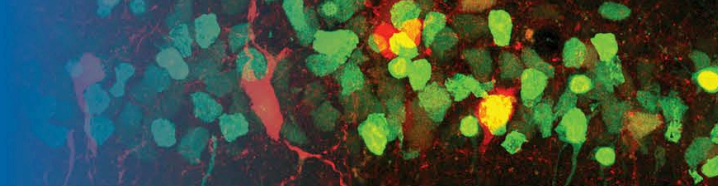
Howard Hughes Medical Institute

Genomic Analysis Laboratory, Salk Institute for Biological Sciences,

San Diego, California

Single-Nucleus DNA Methylation and 3D Genome Organization in Neurotypical and Alzheimer's Brain Cells

Understanding the intricate gene-regulatory mechanisms that dictate the function of diverse cell types is crucial for deciphering the complexities of the brain in both healthy and pathological states. In this study, we conduct a thorough examination of the epigenomes of human brain cells, focusing on single-cell analyses of DNA methylation and chromatin architecture. By integrating data on DNA methylation, chromatin accessibility, structural organization, and gene expression patterns across various cell types and brain regions, we investigate the coordinated changes that underlie cellular function. Moreover, we have pioneered the development of single-cell methylation barcodes. These barcodes leverage the methylation status of specific genomic loci to accurately distinguish between normal and diseased cell types. Our comprehensive, multi-dimensional epigenomic map offers unprecedented insights into the cell-type-specific gene regulatory landscapes present in the adult human brain, shedding light on the molecular underpinnings of neurotypical and Alzheimer's conditions.

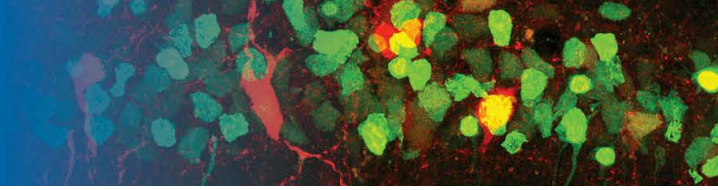


Bing Ren, PhD

Department of Cellular and Molecular Medicine,
University of California, San Diego, La Jolla, California

Single-Cell Transcriptome, Epigenome and 3D Chromatin Architecture Analysis Reveals Age-Associated Gene Regulatory Programs in the Human Hippocampus

Age-associated gene expression changes in the mammalian brain have been observed, yet our understanding of the regulatory mechanisms underlying these alterations and their connection to changes in chromatin structure remains limited. To unravel these complexities, we have applied a comprehensive multi-omics approach, integrating single-cell gene expression, chromatin accessibility, DNA methylation, and 3D chromatin architecture data obtained from the hippocampi of 40 neurotypical human donors spanning ages 20 to 95 years. In general, glia cell types exhibited a significant degree of age-associated transcriptomic and epigenomic changes. We observed a striking correlation between age and loss in the number of astrocytes and oligodendrocyte precursor cells (OPC). Consistent with the loss of these cells, we saw activation of autophagy, apoptotic programs, and senescent marker genes in the cells from older donors. Additionally, we observed a substantial decline in the subset of astrocytes that play a role in maintaining synapses and regulating synaptic transmission. With age, inhibitory neurons exhibited a decrease in expression of genes involved in synaptic vesicle cycle. In astrocytes from older donors, we detected erosion of the 3D genome architecture, where insulation between chromatin domains is diminished, leading to increased aberrant long-range chromatin interactions and differential expression of nearby genes. During aging, microglia undergo a switch from a predominant homeostatic state to an age-associated state having activated inflammatory gene expression. Our data identifies age-associated changes in cell types/states and gene regulatory features that can provide mechanistic insight into the loss of synapses and cognitive decline that occurs in the human brain during aging.



Mariano Gabitto, PhD

Department of Brain Science,

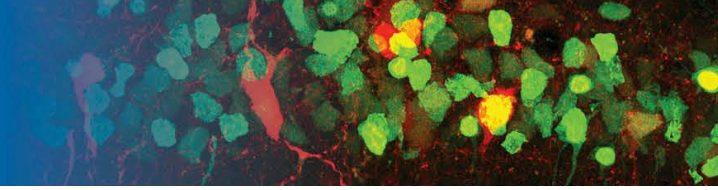
Allen Institute for Brain Science, Seattle, Washington

A multimodal atlas of the molecular and cellular changes to cortex driven by Alzheimer's Disease

Histopathology studies of Alzheimer's disease (AD) have long noted dramatic, progressive, and stereotyped changes across numerous brain regions, including protein aggregation and selective loss of molecularly defined neuronal populations. But the underlying molecular and cellular mechanisms that cause AD and facilitate its progression remain unknown or are only coarsely understood, hampering efforts to treat or cure the disease.

To uncover these mechanisms, we characterized the transcriptomic and epigenetic landscapes of AD from 3 cortical regions (medial entorhinal cortex, middle temporal gyrus, and prefrontal cortex (DLPFC)) by applying single nucleus RNA and ATAC sequencing to ~8 million nuclei isolated in 84 aged donors that span the histopathological and cognitive disease spectrums as part of the broader Seattle AD Brain Cell Atlas (SEA-AD) effort. We leveraged machine learning approaches to hierarchically define ~130 highly resolved transcriptional cell types borrowing from the BRAIN initiatives' neurotypical reference. Next, we utilized Bayesian statistical models to define a continuous scale of pathological progression in each region using multiple measures of pathological proteins (a-Syn, pTDP-43, beta-Amyloid and pTau), and cellular populations. These tools enabled characterization of cell type abundance, gene expression, and chromatin accessibility differences associated with AD.

Our comprehensive molecular atlas identified specific neuronal and non-neuronal subsets, having altered abundances, gene expression, and/or chromatin accessibility, and suggesting they may be selectively vulnerable to disease processes. Their differentially expressed genes and accessible chromatin regions provided new clues to the molecular pathways that underpin AD. We integrated with SEA-AD 10 external datasets that had applied snRNA-seq to 4.3 million cells from the DLPFC of 780 donors. We used this as replication cohorts and identified cellular changes consistently associated with AD. Finally, we created mapping algorithms to map published single cell data sets to our taxonomical reference. These mapping tools create a resource for the research community to align newly profiled cells to a common reference frame. All data sets and mapping algorithms are available at sea-ad.org

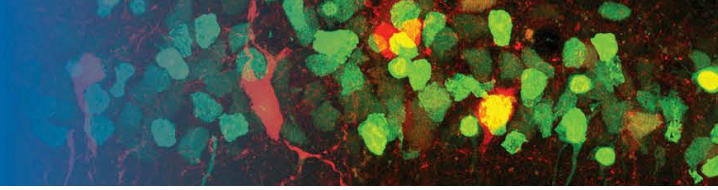


Giorgio Ascoli, PhD

Bioengineering Department & Neuroscience Program,
George Mason University, Fairfax County, Virginia

From neuron types to circuit connectivity: quantitative classification of axonal projections

Axonal morphology is one of the most distinguishing features to identify neuron types. Moreover, the anatomical targeting of axons invading distinct brain areas provides a fundamental blueprint for circuit connectivity. Although anatomists have long recognized the qualitative importance of projecting axons, only recent technological advances enabled the collection of the large datasets required for objective classification. I will present a data-driven approach to quantify axonal patterns by rigorous statistical analysis. Moreover, I will argue that the same methodology is directly applicable to transcriptomic data. Lastly, I will illustrate how this strategy can shed light on functional circuitry.

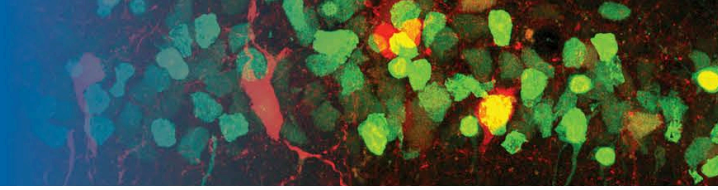


Elly Nedivi, PhD

Department of Brain and Cognitive Sciences,
The Picower Institute for Learning and Memory,
Massachusetts Institute of Technology, Cambridge, Massachusetts

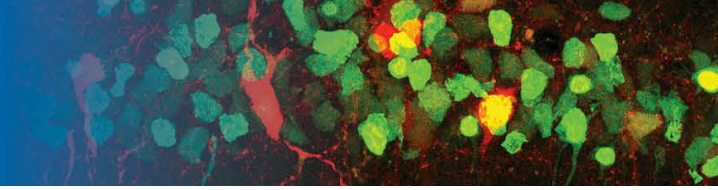
Mapping different excitatory and inhibitory input types onto single cortical pyramidal neurons

The introduction of two-photon microscopy for in vivo imaging has opened the door to chronic monitoring of individual neurons in the adult brain, and the study of synaptic distribution and structural plasticity mechanisms at a very fine scale. We have developed methods for labeling and chronic tracking of excitatory and inhibitory synapses across the dendritic arbors of Layer 2/3 (L2/3) cortical pyramidal neurons in vivo. These methods, combined with posthoc tissue expansion microscopy, have allowed us to experimentally generate synaptic maps of individual L2/3 pyramidal cells of primary visual cortex, revealing the number, density, and size of excitatory and inhibitory synapses onto these neurons as they relate to their afferent source. Based on their afferent identity we can separate excitatory synapses into thalamocortical vs intracortical subpopulations, and inhibitory synapses into somatostatin vs parvalbumin subtypes. Tissue clearing and expansion microscopy of in vivo imaged neurons then allows super- resolution imaging of the pre and postsynaptic features of different synapse types. This work is a first step in defining subtype-specific and proteomic synaptic heterogeneity, an important step towards understanding their differential functionality and distinct roles in neuronal computation and circuit function.



Session 2

Precision Brain Cell Access and Imaging



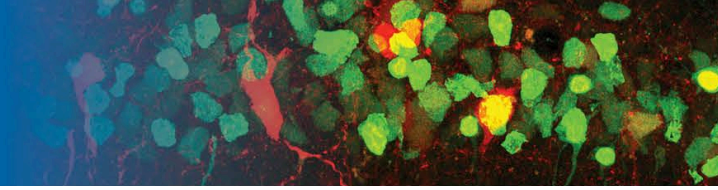
Gordon Fishell, PhD

Department of Neurobiology,

Harvard University, Cambridge, Massachusetts

Identifying enhancers for use in rAAV targeting of specific cortical interneuron subtypes

Work over the past decade has identified that a repertoire of 30 interneuron subtypes exist within cortical circuits and regulate signaling across a breadth of distances, cell types and time scales. While genetic approaches have provided the means to target and manipulate these populations, these approaches require the complex breeding of recombinase Cre and Flp driver lines with appropriate reporters. Not only do these methods require extensive breeding of particular alleles, the appropriate lines are only possible in model genetic species such as mice. To create methods to make inhibitory circuits more accessible across species, including non-human primates, we have used single cell genetics and chromatin methods to identify short enhancers that can be used in the context of AAVs to target and manipulate different inhibitory populations. Although we have yet to identify enhancers for all interneuron subtypes, we have been able to assemble a collection of interneuron-specific AAV constructs to target the major classes of interneuron subtypes in the cortex and hippocampus. Moreover, the majority of these AAVs are able to target these interneuron species across both rodents and primates and can be used to delivery effectors able to identify, activate, track activity or trace circuitry.



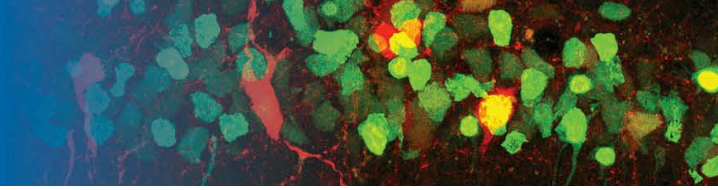
Tanya Daigle, PhD

Department of Human Cell Types,

Allen Institute for Brain Science, Seattle, Washington

Enhancer AAVs for basic and translational neuroscience applications

The brain is the most complex organ in the human body and is comprised of a diversity of cell types that execute a variety of functions. Recent advances in single cell profiling have enabled better definitions of cell types and provided a framework for their systematic characterization. Genetic tools to selectively access these specific cell populations and perturb them in different experimental contexts are critical to further our knowledge of brain function in healthy and diseased states. In this seminar, I will present our enhancer AAV technology platform (Graybuck, Daigle et al., Neuron 2021 and Mich et al., Cell Reports 2021) and share collections of off-the- shelf AAVs that can used in a flexible and cost-effective manner to target one or more cell types and/or brain regions across species, and that can be combined with existing transgenic mouse lines such as our TIGRE-based reporters (Daigle, Madisen et al., Cell 2018). I will discuss the potential of cell type-specific AAVs for treating circuit-based brain disorders and the various challenges in the current CNS human gene therapy space.



Scott Sternson, PhD

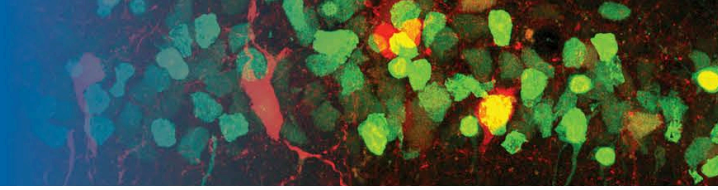
Howard Hughes Medical Institute

Department of Neuroscience,

University of California, San Diego, La Jolla, California

Merging Molecular and Systems Neuroscience with CaRMA Imaging

Brain function is sometimes compared to an orchestral ensemble, where subgroups of neurons that have similar activity are analogous to different types of instruments playing a musical score. Brains are composed of specialized neuronal subtypes that can be efficiently classified by gene expression profiles measured by single-cell RNA sequencing (scRNA-seq). Are these molecularly defined cell types the “instruments” in the neural ensemble? To evaluate the role of molecularly defined cell types in the neural ensemble, it is important to monitor activity in many individual neurons with subsecond temporal resolution along with quantitative gene expression information about each cell. For this, we developed the CaRMA (calcium and RNA multiplexed activity) imaging platform in which deep-brain two-photon calcium imaging of neuron activity is performed in mice during multiple behavioral tasks. This is followed by ex vivo multiplexed RNA fluorescent in situ hybridization to measure gene expression information in the in vivo–imaged neurons. Within a molecularly defined cell type, neurons often showed similar activity patterns such that we could predict functional responses of individual neurons solely from their quantitative gene expression information. Behavioral states could be decoded with high accuracy based on combinatorial assemblies of PVH cell types, which we called “grouped-ensemble coding” akin to the groups of instruments in an orchestral ensemble. Additionally, our data indicate that neuromodulation plays the role of conductor in this ensemble. CaRMA imaging offers a solution to the problem of how to rapidly evaluate the function of the panoply of cell types being uncovered with scRNA-seq. Merging systems and molecular neuroscience is essential to understanding the relationships of gene expression, brain function, behavior, and ultimately neurological diseases.

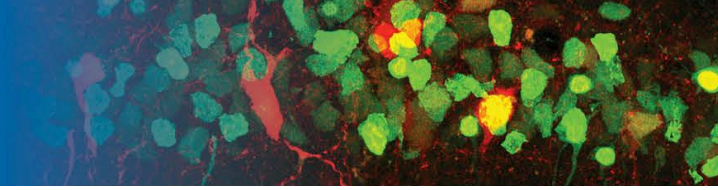


Lin Tian, PhD

Department of Biochemistry and Molecular Medicine,
University of California, Davis, Davis, California

Unlocking neuropeptide dynamics with genetically-encoded biosensors

Neuropeptides are ubiquitous in the nervous system. Research into neuropeptides has been limited by a lack of experimental tools that allow for the precise dissection of their complex and diverse dynamics in a circuit-specific manner. Opioid peptides comprise a clinically relevant family that modulates pain, reward, and aversion. To illuminate the spatiotemporal dynamics of endogenous opioid signaling in the brain, we developed a class of genetically- encoded fluorescent sensors based on kappa, delta, and mu opioid receptors: κLight, δLight, and μLight, respectively. We characterized the pharmacological profiles of these sensors in mammalian cells and in dissociated neurons. We used κLight to identify electrical stimulation parameters that trigger endogenous opioid release and the spatiotemporal scale of dynorphin volume transmission in brain slice. Using in vivo fiber photometry, we demonstrated the utility of these sensors in detecting optogenetically-driven opioid release and observed differential opioid release dynamics in response to fearful and rewarding conditions. I will also discuss development, characterization and applications of other neuropeptide sensors probing broad types of neuropeptides.



Mikhail G. Shapiro, PhD

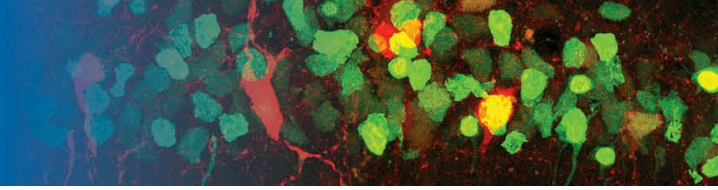
Howard Hughes Medical Institute

Division of Chemistry and Chemical Engineering,

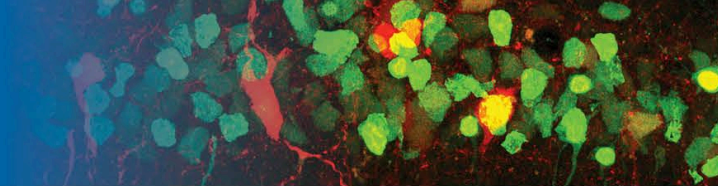
California Institute of Technology, Pasadena, California

Talking to Cells with Ultrasound

To study biological function in intact organisms and develop advanced cellular therapies, we need methods to image and control the function of specific cells deep inside the body. Doing so with predominant optical tools such as fluorescent proteins and optogenetics is challenging due to limited light penetration. In contrast, ultrasound provides deep anatomical access with high spatiotemporal resolution (1 ms and 100 µm, or <10 µm with super-resolution methods). However, historically, ultrasound has not been connected to the function of specific cells. Our work attempts to bridge this gap by engineering biomolecules with the appropriate physical properties to interact with sound waves. In this talk, I will first describe our work with gas vesicles (GVs) – air-filled proteins derived from buoyant photosynthetic microbes – which we are developing as a "GFP for ultrasound". When introduced into cells, GVs scatter sound waves, enabling their detection with ultrasound. By understanding and harnessing their unique biophysical properties, we can use GVs to image a variety of cellular functions in vivo. In addition, under the right ultrasound conditions, GVs can push cells around or disrupt them mechanically with inertial cavitation. Second, I will talk about how we can harness the interactions of ultrasound with moving red blood cells and mechanosensitive ion channels to image and modulate brain activity, creating a foundation for ultrasonic brain-machine interfaces. Finally, I will discuss how ultrasound makes it possible to remote-control therapeutic cells to improve the efficacy and safety of cellular therapies.



Session 3
Neural Circuits and Networks

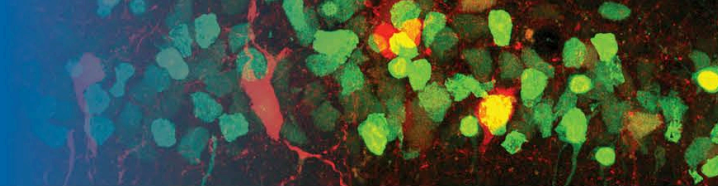


Larry Swanson, PhD

Departments of Biological Sciences and Psychology,
University of Southern California, Los Angeles, California

A model of the brain's internal network organization: a conceptual framework for systems neuroscience research

Current evidence suggests that the bilateral mammalian nervous system has about 1,000 gray matter regions, a million possible axonal connections between them (1,000 2), and 2 1,000 possible combinations of connections. Network neuroscience tools are used here to clarify the basic plan of this complex bodily system, which coordinates external behavior with internal homeostasis. In the strategy we are developing, two fundamental tools are cluster analysis of male and female axonal connection datasets (rat), along with a guiding hypothesis that sets of strongly connected regions form structure-function subsystems or modules. The results suggest a model with a nested hierarchy of identified structure-function subsystems that are weakly interconnected. The usefulness of this model as an analysis tool was tested by computationally "lesioning" (removing) each of the 279 right-left region pairs in the forebrain-midbrain network and quantifying the effect on overall network organization. The results indicate that focal lesion effects (and presumably other types of effects) tend to concentrate, "spread," or constrain within a set of related subsystems previously identified by cluster analysis. Furthermore, the composition of the subsystem set is influenced by the set of input-output connections associated with the lesioned region, and as expected, the magnitude of network effects is positively correlated with the lesioned region's network centrality. These and other results suggest a general model of the nervous system based on a nested hierarchy of identified structure-function subsystems or modules that are themselves weakly interconnected. Furthermore, the underlying dataset can serve as a framework for online experimental, clinical, theoretical, computer science, and educational applications.



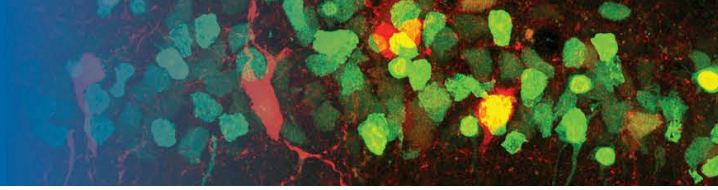
Hong-Wei Dong, MD, PhD

Department of Neurobiology,

University of California, Los Angeles, Los Angeles, California

Neural Networks of the Cortico-Basal Ganglia-Thalamic System

The cortico-basal ganglia-thalamic-cortical system is one of the fundamental network motifs in the brain, essential for cognitive functions, sensorimotor integration, and the development of various neurological and psychiatric disorders. Over the past decade, pioneering research from our team and others around the world has led to significant breakthroughs in mapping the complex structural and neural circuits frameworks of this key network. In my upcoming presentation, I will highlight these sophisticated models of the network's architecture and explore how these anatomical discoveries offer promising new directions for research in functional and behavioral neuroscience.

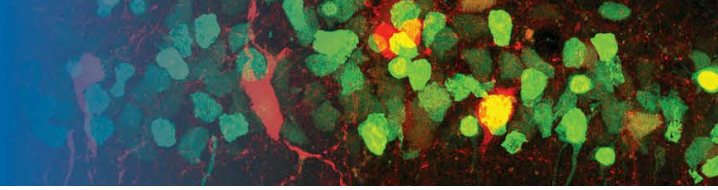


Li Zhang, PhD

Department of Physiology & Neuroscience,
University of Southern California, Los Angeles, California

Excitation/Inhibition Imbalance of MPOA Circuits for Stress-Induced Depressive-Like States

The etiology of depression is known to be diverse, and different etiological factors can affect neural circuits differently although leading to common manifestations of behavioral deficits in animals. Whether there is a universal principle governing the expression of depressive states is not well-understood. Dysregulation of E-I balance has been previously postulated to contribute to a range of neuropsychiatric diseases including depression. However, in which specific brain structure(s) or neural pathways the E/I balance is dysregulated, and how the maladaptation of E/I balance can result in depressive-like states remain largely unclear. I will discuss our recent studies on the imbalance of excitation-inhibition in hypothalamus-midbrain circuits as a potentially general neural circuitry mechanism for depressive states induced by different etiological factors.

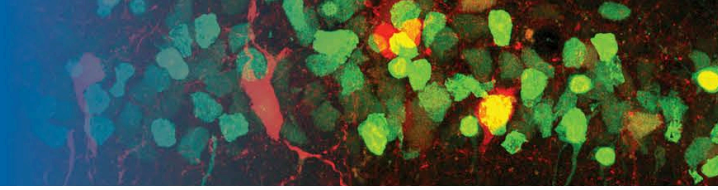


Wei Wei, PhD

Department of Neurobiology, University of Chicago, Chicago, Illinois

Context-dependent motion processing by the retinal circuitry

How sensory circuits adaptively adjust their input-output relationships according to the sensory inputs is ethologically important but poorly understood. Our recent work aims to determine the contextual modulation of retinal circuit function by visual features frequently encountered by the animal in the natural environment. In this talk, I will present our findings on context-dependent motion processing in the mouse retina at multiple levels: from recruitment of different sets of mechanistic components, to algorithmic changes of circuit motifs, to the dynamic encoding of multiple visual features.

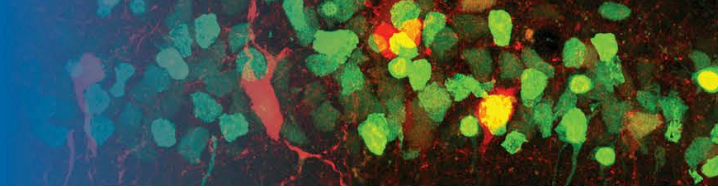


Hysell Oviedo, PhD

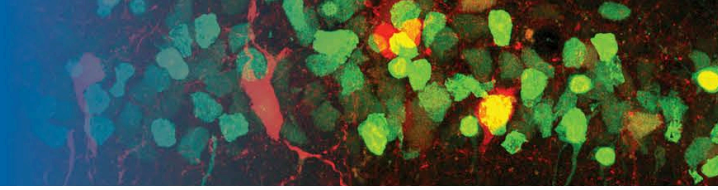
Department of Neuroscience, Washington University, St. Louis, Missouri

Asynchronous Development of the Mouse Auditory Cortex in the Left and Right Hemispheres

A fundamental feature of mature auditory sensory processing is the allocation of specialized cognitive functions to the Left and Right Auditory Cortex (ACx). Despite the significant role of lateralized auditory processing in human cognition, our understanding of the developmental mechanisms that underlie this cortical specialization remains limited. To bridge this gap in knowledge, our objective is to conduct a comparative analysis of Left and Right ACx functionality in mice across different developmental stages. Interestingly, our recent findings reveal that cellular and network signs of maturity in the Auditory Cortex appear earlier in the right hemisphere in mice. Additionally, we demonstrate that persistent, experience-dependent map reorganization is confined to the hemisphere undergoing active maturation and can be differentially engaged by temporally limited manipulations of the sensory environment. These findings suggest that differential timing in hemisphere development could lead to lateralized auditory functioning.



Session 4
Brain Disorders

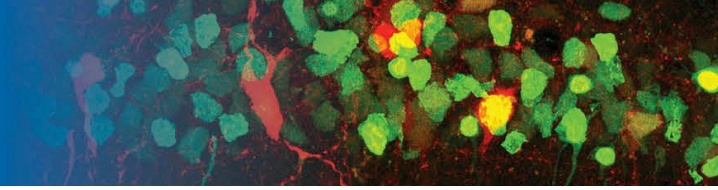


Xiaoke Chen, PhD

Department of Biology, Stanford University, Stanford, California

A closed-loop circuits for chronic pain

Inflammation or nerve injury at periphery can cause chronic pain. The spinal cord-projecting neurons in the rostral ventromedial medulla (RVMSC neurons) play active roles in pain facilitation. However, the neuronal pathway that transmitting nociceptive information from periphery to these descending RVMSC neurons is unknown. Here we report a closed-loop circuit extending from spinal cord to ventral posterolateral thalamus, proceeding to primary somatosensory cortex; then returning to the spinal cord via lateral superior colliculus, ultimately connecting to m-opioid receptor expressing RVMSC neurons. Silencing any node along this multisynaptic circuit has minimal effect on nociception in healthy mice, but it eliminates mechanical hypersensitivity and restores normal nociceptive response thresholds in mouse models of inflammatory and neuropathic pain. Repetitive, but not acute, activation each node within this circuit in healthy mice is sufficient to cause robust chronic mechanical hypersensitivity. Our findings elucidate a spinal-brain-spinal circuit loop linking ascending and descending pathways that specifically drives chronic mechanical pain, and identify novel circuitry targets for treating chronic pain.



X. William Yang, MD, PhD

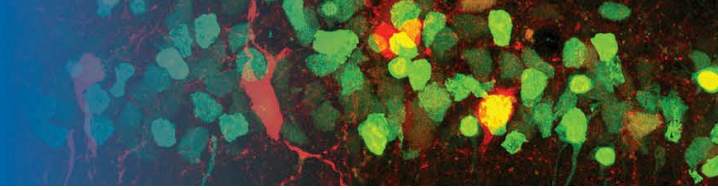
Center for Neurobehavioral Genetics

Department of Psychiatry & Biobehavioral Sciences,

University of California, Los Angeles, Los Angeles, California

Genetic Drivers of Selective Neuronal Pathogenesis in Huntington's Disease Mice

DNA mismatch repair (MMR) genes are among the GWAS modifiers of Huntington's disease (HD) and can modify somatic mutant Huntingtin (mHtt) CAG-repeat instability. However, the precise roles and effect sizes of MMR/GWAS genes on HD pathogenic processes beyond repeat instability remain unclear. Here we tested knockout (KO) alleles for nine HD modifier/MMR genes in mHtt knockin (mHtt-KI) mice to probe disease-modifying mechanisms. KO mice for four MMR genes nearly fully or partially rescued early-onset triad of striatal medium-spiny-neuron (MSN) selective phenotypes: somatic CAG-repeat expansion, transcriptomic dysregulation, and mHtt aggregation. Other canonical MMR genes and HD GWAS genes did not show modifier effects. Mechanistically, purified mHtt-KI MSNs (but not non-neuronal cells) exhibited population-scale CAG-repeat migration, which is necessary for pathogenic onset; and such expansion is slowed or stopped with MMR mutants. Critically, mHtt-KI mice exhibited late-onset cortical neuronal pathogenesis characterized by similar pathogenic triad as in the striatum, and MMR mutants also prevented the cortical pathology. Moreover, MMR mutants reversed synaptic marker loss, glial activation, and locomotor deficits in aged HD mice. Thus, a subset of MMR genes are genetic drivers of selective neuronal pathogenesis in both the striatum and cortex of HD mice, providing novel mechanistic insights and in vivo platforms for targeted HD therapeutic development.

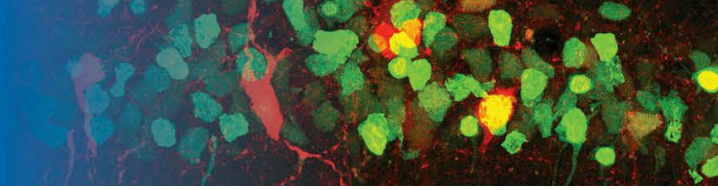


Nicholas Seyfried, PhD

Department of Biochemistry, Emory University, Atlanta, Georgia

Integrated Proteomics for Novel Target and Biomarker Discovery in Alzheimer's Disease

The proteome is the "executioner" of the effects of aging, genetics, the environment, and other risk factors that cause human disease. In Alzheimer's disease (AD), amyloid-beta (Aβ) and Tau were identified with classical biochemical methods, enabling breakthroughs from discovery as proteins directly altered in AD pathology. We hypothesize the AD proteome undoubtedly contains many other proteins that play key roles in the initiation and progression of AD. To further our understanding of AD pathophysiology and to identify new targets for early intervention, we employed unbiased large-scale quantitative proteomic methods utilizing high- resolution mass spectrometry platforms. To date, we have analyzed over 2000 individual brain tissues from multiple regions, quantifying nearly 9,000 proteins per sample. Systems biology approaches, including Weighted Gene Co-expression Network Analysis (WGCNA), have enabled us to resolve modules or communities of proteins that correlate strongly with clinical and pathological AD phenotypes. These modules reflect key mechanisms linked to impaired synaptic, vascular, and glial function. These comprehensive brain proteomic datasets linked to AD and other neurodegenerative diseases are then integrated with the cerebrospinal fluid (CSF) and plasma proteome profiles in AD patients to prioritize brain-based biomarkers in humans for the diagnosis, staging and therapeutic response to treatment.

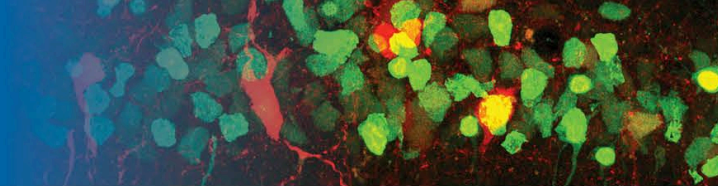


Monica Carson, PhD

Department of Biomedical Sciences,
University of California, Riverside, Riverside, California

Lung-brain connection: Sex- and age-specific neuroinflammatory responses to systemic inflammation

While the brain is often viewed as being isolated and protected from systemic insults by the blood-brain-barrier (BBB), systemic inflammation triggered by fungal, bacterial and viral components trigger region, sex- and age-dependent specific responses within the brain. Based on profiling gene expression and histologic analysis, we quantify the differential consequences of airborne exposure of non-infectious Alternaria alternata fungal particulates or LPS (gram negative bacterial component). We find that exposures sufficient to cause lung inflammation also cause sex-specific effects in neuroinflammatory, microglial and synaptic responses in the brainstem region regulating respiration, and in the hippocampus.

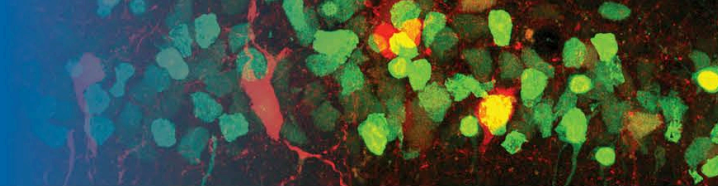


Kalpna Gupta, PhD

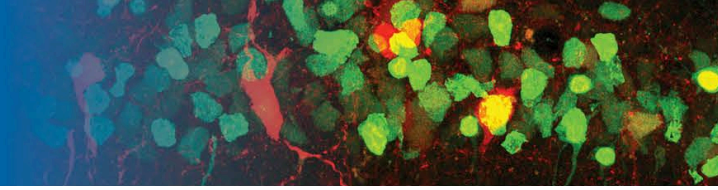
Department of Medicine, University of California, Irvine, Irvine, California

Pain at the intersection of social and neural networks

Environment influences biology in many ways through, chemical, organismal and perception-based stimuli. As compared to chemical and infection-induced impact on cell and molecular biology, modeling perception-based neural cellular and molecular interactions remain understudied. We developed a model of isolation and companionship-induced influence on neural networks and the mechanisms of interoception. Using a sickle cell disease (SCD) mouse model and wild type mice we examined the mechanisms of interoception and neural networks related to pain under a naturally mimicking impact of social isolation. Male sickle and control mice were placed under 'isolation' housed one to a cage and in parallel for companionship, a male mouse was placed with a female in the same cage for a period of 1-3 months. We observed an increase in thermal and mechanical hyperalgesia and impairment in cognitive function in sickle mice under isolation compared to those with a companion. MERFISH analysis of brain showed molecular alterations at a multicellular level in the brain. These neural signals converged at an increase in calmodulin kinase 2A, the central calcium signaling which has a detrimental impact on pain. Additionally, the neural network alterations were accompanied by an alteration in inflammation in the periphery. In conclusion, perception influences neural networks in a 2-way communication between the central nervous system and the peripheral mechanisms.



Session 5
Frontiers, New Concepts and Approaches

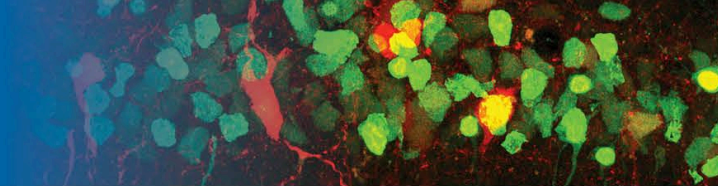


Jianhua Cang, PhD

Departments of Biology and Psychology,
University of Virginia, Charlottesville, Virginia

Mapping Visual Functions onto Molecular Cell Types in the Superior Colliculus

The superior colliculus (SC) is an evolutionarily conserved structure that receives direct retinal input in all vertebrates. It was the most sophisticated visual center until the neocortex evolved in mammals. Even in mice and tree shrews, mammalian species that are increasingly used in vision research, the vast majority of retinal ganglion cells project to the SC, making it a prominent visual structure in these animals. In this talk, I will review our recent functional studies of the mouse SC and describe our current efforts in linking functional properties to genetically identified cell types in both mice and tree shrews.



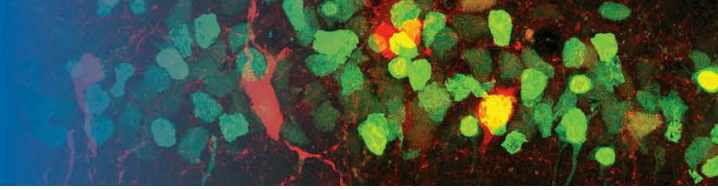
Joshua Trachtenberg, PhD

Department of Neurobiology,

University of California, Los Angeles, Los Angeles, California

The early evolution of cortical representation and coding

The emergence of mammals in the Late Triassic, some 225 million years ago, brought with it new features, including endothermy, hair, viviparity, lactation, a four chambered heart, and three middle ear bones. Perhaps most profound was the emergence of the six layered neocortex. Neocortex has expanded considerably thereafter from marsupials, where it occupies less than 20% of the brain to primates, where cortex is 80% of brain mass. The shift in cortical organization from glires (rodents and lagamorphs) to primates has been extensively studied. Much less is known of the transition in cortical organization as mammals evolved from marsupials to placentals. Here, we examine the molecular cell types in the south american opossum and compare them to those in mice. While cortical excitatory cell types are stable across this transition, we find a large increase in the ratio of upper layer inhibitory neurons derived from the caudal ganglionic eminence with a concomitant reduction in fast-spiking, parvalbumin positive interneurons. Using chemogenetics to regress the mouse visual cortex towards a more opossum-like state (reduced upper layer inhibition with enhanced PV inhibition), we examine the impact of this inhibitory shift on cortical computation. Our overarching hypothesis is that this shift enhanced cortical representation and coding and promoted fitness.

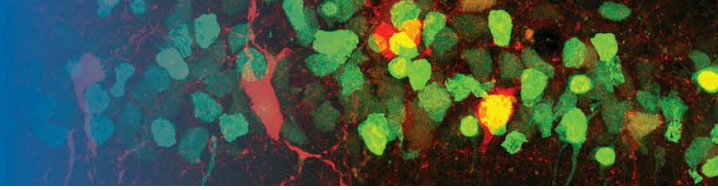


Gianluca Tosini, PhD

Department of Pharmacology & Toxicology,
Morehouse School of Medicine, Atlanta, Georgia

Regulation of visual processing and photoreceptors' viability by the circadian Clock

The retinal circadian clock was the first extra-SCN circadian oscillator to be discovered in mammals. Several studies have now established that many aspects of mammalian retinal physiology is under the control of a retinal circadian clock. Within the retina melatonin release, dopamine synthesis, gamma-aminobutyric acid (GABA) turnover rate and release, extracellular pH, rod disk shedding and photopigments gene expression, are all regulated in a rhythmic manner. In addition, circadian signals originating in the retina drive rhythms in the hypothalamic SCN clock, even in the absence of light/dark cycles. Additional experimental evidence also suggests that other ocular structures (e.g., retinal pigment epithelium and cornea) possess functional circadian clocks. The gene Bmal1 is a key component of the mammalian circadian clock and a few studies have investigated the role of Bmal1 in the eye. Removal of Bmal1 from the retina abolishes the circadian rhythm in the photopic electroretinogram and also changes the spatial expression of cone opsins. Additional studies have also revealed that removal of this clock gene from the retina significantly affects visual information processing in both rod and cone pathways, reduces the thickness of inner retinal nuclear and plexiform layers, accelerates the decline of visual functions during aging, and reduces the viability of cone photoreceptors. Finally, removal of Bmal1 from the retinal pigment epithelium - but not from the retina - abolishes that circadian rhythm in disk mice. Thus, it appears that dysfunction in the retinal circadian clock, caused by genetic or other means, may play an important role in the decline of visual function during development and/or aging.

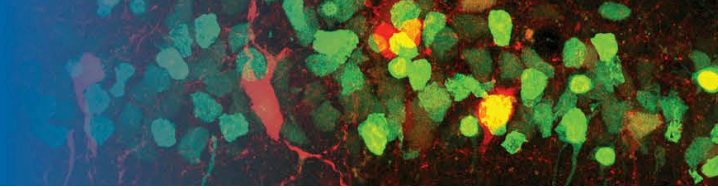


Xiaoning Bi, MD, PhD

College of Osteopathic Medicine of the Pacific,
Western University, Pomona, California

Lysosome-mediated regulation of brain development and learning and memory: Implications for Angelman syndrome

Accumulating evidence has established lysosomes as a critical organelle regulating multiple cellular functions, including energy sensing, through the regulation of mechanistic target of rapamycin complex 1 (mTORC1), and intracellular calcium homeostatic regulation, via multiple lysosomal cation channels. Our recent research has shown that both processes are implicated in the pathogenesis of Angelman syndrome (AS), a rare neurodevelopmental disease caused by the deficiency of the maternally inherited ubiquitin E3 ligase, UBE3A, which mediates polyubiquitination of protein substrates. We showed that LAMTOR1, a member of the Ragulator complex, which is essential for full activation of mTORC1, is a substrate of UBE3A. Ube3a ubiquinates LAMTOR1, resulting in its proteasomal degradation, and Ube3a deficiency in AS mice, which lack maternal Ube3a, induces increased lysosomal localization of the Ragulator and activation of mTORC1. LAMTOR1 knockdown in hippocampal CA1 neurons of AS mice reduces elevated mTORC1 activity and improves dendritic spine maturation, long- term potentiation (LTP), as well as learning performance. We have also shown that LAMTOR1 regulates the transient receptor potential mucolipin 1 (TRPML1) channel which mediates lysosomal Ca^{2+} release, independently of mTORC1. LAMTOR1 tonically inhibits TRPML1-mediated Ca^{2+} release, thereby regulating dendritic lysosomal motility, synaptic plasticity, and learning. We further showed that TRPML1 activation stimulates exosome release from wildtype (WT) synaptosomes, but not from AS synaptosomes, possibly due to increased inhibition of TRPML1 by LAMTOR1. Treatment with exosomes from WT neurons improved dendritic spine development and maturation and learning performance in AS mice. Altogether, our results identify novel mechanisms through which lysosomes regulate synaptic development and plasticity and learning and memory.

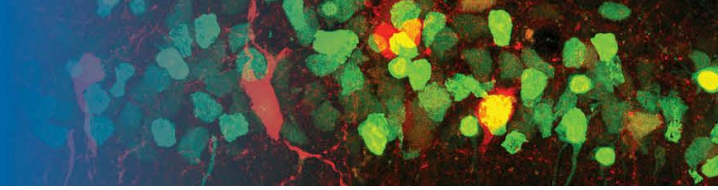


Michael Koob, PhD

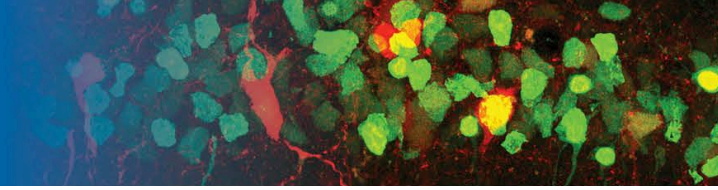
Department of Laboratory Medicine & Pathology,
University of Minnesota, Minneapolis, Minnesota

Modeling the genetics of human dementias in mice: full human gene-replacement mouse models of Alzheimer's Disease and Related Dementias

Genetic studies conducted over the past four decades have provided us with a detailed catalog of genes that play critical roles in the etiology of Alzheimer's Disease (AD) and AD Related Dementias (ADRDs). Despite this progress, as a field we have had only limited success in incorporating this rich complexity of human AD genetics findings into our animal models of this disease. Our primary goal for the Gene Replacement-AD (GR-AD) project is to develop mouse lines that model the genetics of AD as closely as possible. To do this, we are generating mouse lines in which the genes-of-interest are precisely and completely replaced in the mouse genome by their human orthologs. Each Gene Replacement (GR) model set consists of a control line with a wild type human allele, and variant lines that precisely match the human genomic sequence in the control line except for a high-impact pathogenic mutation or risk variant that we specifically introduce. These precisely matched GR sets of animal models allow the research community to evaluate the molecular impact of pathogenic mutations and risk variants within the context of the human genomic sequence in which they occur in patients, and these mouse lines contain all potential human therapeutic targets ranging from the full genomic DNA sequences to all RNA transcription variants and protein products that they encode.



Session 6
TBD and Viral Core Workshops



Waldo Cerpa, PhD

Department of Biological Sciences,

Pontificia Universidad Católica de Chile, Santiago, Chile

Unveiling the Role of NMDA Receptors in Traumatic Brain Injury: Distribution, Signaling, and Potential Therapeutic Targets

Traumatic brain injury (TBI) triggers neuronal death through various molecular pathways. One key player is excessive stimulation of N-methyl-D-aspartate receptors (NMDARs) by glutamate, leading to excitotoxicity and activation of cell death cascades. However, our understanding of how NMDAR distribution and signaling contribute to TBI remains incomplete.

Calcium influx through extrasynaptic NMDARs can overload mitochondria, which is crucial for ATP production and calcium homeostasis. Disruption of this balance is linked to neuronal dysfunction and death.

TBI damages the central nervous system (CNS) through multiple mechanisms, including synaptic dysfunction, protein aggregation, mitochondrial dysfunction, oxidative stress, and neuroinflammation. Glial cells, the most abundant cell type in the CNS, play a central role in the brain's response to TBI. Astrocytes, a specific type of glial cell, are critical for maintaining ion balance, energy metabolism, blood-brain barrier integrity, and immune response.

These cellular mechanisms, particularly mitochondrial alterations and glial involvement, provide a framework for understanding how TBI affects the distribution, signaling, and trafficking of NMDA receptors to the cell membrane.

Our research proposes a critical role for STEP61 (Striatal-Enriched protein tyrosine phosphatase) in regulating the phosphorylation state of the GluN2B subunit of the NMDAR. Additionally, we explore the role of exocyst complex proteins like Exo70, which are involved in the constitutive trafficking of glutamate receptors towards synapses. We observed increased exocyst complex assembly and its interaction with GluN2B under TBI conditions.

Identifying these novel players and specific cellular signals during TBI is crucial for developing potential diagnostic tools (early biomarkers) and therapeutic strategies for this debilitating condition.

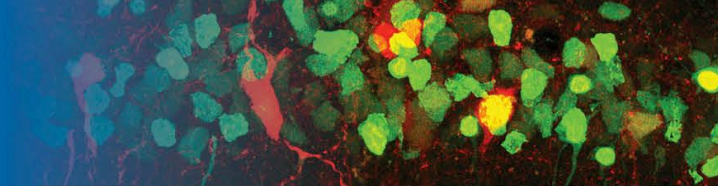
María-Paz Marzolo, PhD

Department of Biological Sciences,

Pontificia Universidad Católica de Chile, Santiago, Chile

ApoER2 trafficking and Reelin signaling defects in neurons defective in the AP-4 adaptor complex: Implications for Hereditary Spastic Paraplegia

Adaptor protein complex 4 (AP-4) is a heterotetrameric complex that promotes protein export from the trans-Golgi network. Mutations in each of the AP-4 subunits cause a complicated form of Hereditary Spastic Paraplegia (HSP). We describe that ApoER2, is a new cargo of the AP-4 complex, identifying the motif ISSF/Y within the ApoER2 cytosolic domain as necessary for interaction with the canonical signal-binding pocket of the μ4 subunit of AP-4. Hippocampal neurons from Ap4e1-KO mice and AP4M1-KO human iPSC-derived cortical i3Neurons exhibit significantly reduced ApoER2 protein expression. Analyses of biosynthetic transport of ApoER2 reveal differential post-Golgi trafficking of the receptor, with lower axonal distribution in KO compared to wild-type neurons, indicating that the interaction of ApoER2 with regulated the axonal localization signaling in mouse hippocampal and human cortical KO neurons. AP-4 deficiency was associated to a reduction of Reelin-induced ERK phosphorylation, CREB activation, and Golgi deployment. Altogether, this work establishes ApoER2 as a novel cargo of the AP-4 complex, suggesting that defects in the trafficking of this receptor and in the Reelin signaling pathway could contribute to the pathogenesis of HSP caused by mutations in AP-4 subunits.



Jorge M Campusano. PhD

Department of Biological Sciences,

Pontificia Universidad Católica de Chile, Santiago, Chile

Tranglutaminase enzyme contributes to brain development and function

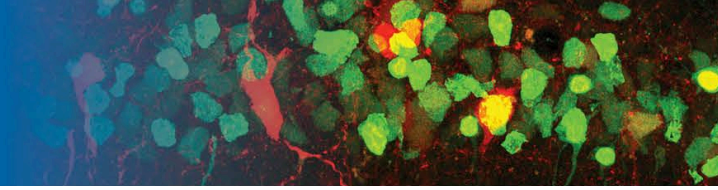
Transglutaminase 2 (TG2), a member of the transglutaminase family, plays a key role in several disorders including cancer progression, celiac and cardiovascular diseases, among other conditions. It has been also suggested that TG2 contributes to several brain disorders including neurodegenerative diseases and CNS injury, due to its ability to crosslink proteins into insoluble aggregates. Although recent reports support that TG2 plays a role in neural development, this is still under investigation. In the Drosophila melanogaster genome there is only one transglutaminase (TG) gene which might be responsible for all actions played by this enzyme family in other animals. Here we have asked whether flies deficient in TG expression exhibit behavioral phenotypes which could be linked to CNS structural alterations.

We studied behavioral phenotypes in flies globally deficient in TG expression - TG d01144 and TG-CRIMIC mutants - or bearing a brain-specific deficiency for the TG enzyme.

Our data show that TG homozygous mutant animals exhibit reduced lifespan as compared to heterozygous flies. Behavioral studies show impaired startle- induced climbing behavior in TG mutant animals. Interestingly, when basal locomotion is studied in an open field test, no motor phenotype is observed although flies exhibit increased centrophobism, which reflects anxiety. Additionally, the TG mutant flies exhibit reduced interaction time with their peers, which suggest asocial behavior.

These behavioral phenotypes are accompanied by alterations in fly brain anatomy, evidenced by structural changes in the mushroom bodies, an important association area in the fly brain, and dopaminergic neurons.

Our data support that TG is a relevant enzyme in brain formation and function.



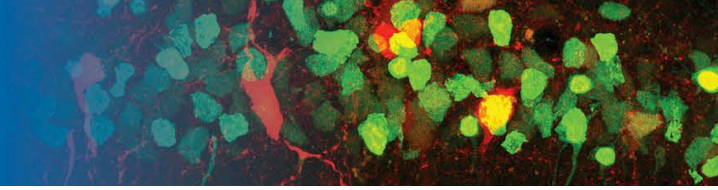
Tim Shay, PhD

Scientific Director of the Clover Center,

California Institute of Technology, Pasadena, California

Reverse engineering mechanisms of blood-brain barrier transcytosis engaged by AAVs during directed evolution

In recent years, diverse groups across academia and industry have used mechanism-agnostic directed evolution screens to populate a broad panel of chemically distinct AAV capsids with greatly enhanced blood-brain barrier (BBB) crossing in model organisms from mouse to macaque. However, their application in nontraditional model organisms and ultimate therapeutic utility in humans is difficult to predict without knowledge of their underlying mechanisms. By applying novel screening approaches, we have identified several previously unreported BBB transcytosis receptors engaged by lab-evolved AAVs. These receptors unlock direct development of species-appropriate brain-penetrant AAVs and, potentially, other therapeutic modalities.



Alexis Bouin, PhD

Center for Neural Circuit Mapping, University of California, Irvine, Irvine, California

Design, production and use of recombinant pseudotyped rabies virus for neural circuit mapping

New viral tools are critical for improving anatomical mapping and functional studies of cell-type-specific and circuit-specific neural networks in the intact brain. The goal of the CNCM viral core is to develop new and improved viral tools that can be used for a broad range of applications and to make them widely available in the neuroscience field. Using the genome of the rabies vaccine strain, SAD B19, we developed multiple recombinant rabies viruses (RV) that encode bright fluorescent proteins along with subcellular localization signals that replace the viral glycoprotein (G) as well as non- fluorescent reporters designed for multi-modal imaging.

We have generated a series of novel spectrally distinguishable recombinant RV that express fluorescent reporters ranging from blue to far-red signal. Among the fluorescent reporters, mNeonGreen and tdTomato are brighter, longer-lasting than others previously described, and they do not quench as easily compared to traditional GFP and RFP (DsRed and mCherry). We also engineered a range of reporters that localize to different sub-cellular compartments, including the cytoplasm, nucleus, cellular membranes, mitochondria, somatodendritic, and pre- and post-synaptic locations. Using a correlated light, X-ray, and electron microscopy workflow (CLXEM) we were able to detect electron microscopy reporters scattered within axons and synaptic termini, as well as higher density near mitochondria of presynaptic boutons.

In summary, our newly developed recombinant RVs offer a range of sub-cellular targeted reporters that facilitate a wide repertoire of cell-type specific and circuit specific mapping studies that are amenable to automated computer analysis. The recombinant RV expressing ferritin allows detection using electron microscopy. Ongoing studies are aimed at attenuating viral toxicity in vivo.