Report of the June 2, 2016 Neuro-Workshop

The Neuro-Workshop was held June 2, 2016 at LBNL with attendees from:

PNNL	Pacific Northwest National Laboratory
LBNL	Lawrence Berkeley National Laboratory
EMSL	Environmental Molecular Sciences Laboratory, a DOE Office of Science User Facility at PNNL
JGI	Joint Genome Institute, a DOE Office of Science User Facility at LBNL
JBEI	Joint Biological Energy Institute at LBNL

Co-chaired by Andrew Wyrobek and Kristofer Bouchard
Report submitted on August 11, 2016

Workshop Report Outline

Executive Summary

The Biosciences Neuro-Workshop was held at the Lawrence Berkeley National Laboratory on June 2, 2016 with participants from two national laboratories (LBNL and PNNL), two BER/DOE User Facilities (EMSL and JGI) and JBEI. This workshop was convened to discuss the relevant technological capabilities for BRAIN and collaborative opportunities to address neuroscience 'grand challenges" in support of the DOE contribution to BRAIN and other National scientific challenges. This workshop builds off previous salons and workshops focused on technological challenges of BRAIN and how National Laboratories and DOE User Facilities can uniquely contribute to addressing these challenges. In the context of scientific problems important to DOE, the workshop addressed the impact of neurotechnologies and neuroscience for determining the serendipitous and deleterious impacts of byproducts of biosynthetic processes and changing environments, enhancing soil and plant health, and formulating predictive understandings of ecosystems.

The White House announced the national BRAIN Initiative in 2013 with the aim to reduce the costs of brain disorders to the US economy through the development of experimental technologies and computational tools to understand brain function across multiple temporal and spatial scales. In March 2016, the White House announced that DOE joined the BRAIN Initiative with a focus on development, characterization, and validation of multi-scale tools required to obtain a dynamic read-out of neurological function.

Technological developments for BRAIN will provide opportunities to advance basic knowledge in environment, bioenergy and other national biomedical imperatives (e.g. the National Microbiome Initiative) by building tools to measure and manipulate large numbers of biological processes simultaneously, and by developing computing methods to extract understanding from the data. This workshop highlighted numerous challenges for BRAIN for which DOE could contribute, ranging from: tools to record activity from many thousands of neurons in behaving animals; genetic identification of neuronal subtypes for subregional sensing and manipulation; imaging tissue microarchitecture; measurements of cerebral spinal fluids; and design of consistent analytic pipelines for multi-modal data integration. The workshop participants agreed that technology development could be accelerated by closing the 'design-test-learn' loop using complementary rodent and insect model systems.

The workshop discussion on technological challenges highlighted broad consensus for synergistic opportunities arising from collaboration across institutes. It was thought that, together, there are at present sufficient capabilities presently to begin multi-scale investigations of nervous system function. Such an investigation would require integration of extant and developing capabilities to: measure neural activity during behavior; use protein engineering to improve and innovate biological markers/sensors/actuators; genetically target specific cell types with said constructs; utilize microfluidic systems for high-throughput cellular screening; perform deep molecular analyses of brain regions and fluids; and investigate bidirectional interactions between environments and organisms. These capabilities would provide a foundation to address major BRAIN challenges of linking the activity of individual and small populations of neurons to the large-scale patterns of neural activity that give rise to sensations and behaviors in living animals, and how they are altered in neurological disease states. For BER mission areas, these tools and pipelines could be adapted to enhance our understanding of how brain function and behavior vary among healthy individuals and how they are changed after exposure to environmental challenges or alterations in host microbiomes, how metazoan behavior effects soil and plant health, and lead to improved approaches to nutrient/microbe monitoring in soil. Such an integrated, multi-scale approach would provide impactful resources for neuroscience researchers interested in understanding brain function from molecules to minds, as well as the broader biology community for determining how molecular and microscopic mechanisms can lead to macroscopic phenomena in complex systems of heterogeneously interacting constituents (e.g. soil-plant and microbe-biome systems).

Neuro-Workshop Program

Introduction

9:30 – 9:50	Introductions and Meeting Goals – Andy Wyrobek	LBNL
9:50 – 10:10	DOE and the BRAIN Initiative – Peter Denes	LBNL

Overviews of Institute Capabilities for the Brain

Moderator: Andy Wyrobek

10:10 – 10:30	EMSL Capabilities - Harvey Bolton	EMSL
10:30 – 10:50	JGI Capabilities – Axel Visel	JGI
10:50 – 11:05	Break - Discuss DOE and BRAIN	Group
11:05 – 11:25	JBEI Capabilities – Blake Simmons	JBEI
11:25 – 11:45	LBNL Capabilities – Kris Bouchard	LBNL
11:45 - 12:30	Working lunch - Complementarity of Institute Capabilities.	Group

Short Talks - Brain Science Vision

Moderator: Kris Bouchard

12:30 – 12:35	Statement of Goals - Moderator	LBNL
12:35 – 12:50	Janet Jansson, Microbiomes and BRAIN	EMSL
12:50 – 1:05	Sue Celniker, Genomic Response to Environment	LBNL
1:05 – 1:20	Axel Visel, Mouse Studies	LBNL
1:20 – 1:35	Andy Wyrobek, Diversity for Neurological Risks	LBNL
1:35 – 1:50	Hector Garcia Martin, Fluxes as Predictors of Function	JBEI
1:50 – 2:05	Diane Dickel, Single-Cell Capabilities for BRAIN	LBNL
2:05 – 2:20	Javier Ceja-Navarro, Ecosystem Dependence on Metazoan Behavior	LBNL
2:20 – 2:35	Steve Wiley, EMSL and PNNL Vision for Neurobiology	EMSL
2:35 – 2:50	Cynthia McMurray, Neurodegeneration and Movement Disorder	LBNL
2:50 – 3:00	Break - Discuss Presentations	Group

Open Discussion of Big Challenges and Opportunities for Collaboration

Moderators: Peter Denes and Blake Simmons

3:00 – 3:10	Statement of Goals - moderators	
3:10 – 3:40	Axel Visel	JGI
3:40 – 4:10	Sue Celniker	LBNL
4:10 – 4:40	Steven Wiley	EMSL

Workshop Summary Planning

Moderators: Andy Wyrobek and Kris Bouchard

4:40 -5:20	Institute POCs (alternate)	
	Axel Visel (Diane Dickel)	JGI
	Blake Simmons (Hector Garcia Martin)	JBEI
	Harvey Bolton (Steve Wiley)	EMSL
	Andy Wyrobek and Kris Bouchard	LBNL

Group dinner at local Berkeley Restaurant

5:20 – 9:00	Discuss complementarity of capabilities and opportunities for communication among institutes and opportunities for collaborations.

Workshop Motivation and Goals

The June 2nd Neuro-workshop followed previous salons, summits, and workshops on the technological challenges of the BRAIN Initiative and how DOE National Laboratories and DOE User Facilities can uniquely contribute to these challenges. The Presidential BRAIN Initiative was announced in April 2013 as "a bold new Grand Challenge focused on revolutionizing our understanding of the human brain." With the initial participation of NIH, NSF, and DARPA, the "BRAIN Initiative has the potential to do for neuroscience what the Human Genome Project did for genomics by supporting the development and application of innovative technologies that can create a dynamic understanding of brain function. It aims to help researchers uncover the mysteries of brain disorders, such as Alzheimer's and Parkinson's diseases, depression, and traumatic brain injury (TBI)." The NIH has established a roadmap for BRAIN, centered around seven 'Grand Challenge' goals, described in the BRAIN 2025 report. In 2014, FDA and IARPA joined the BRAIN Initiative. A *Summit* in January 2015, sponsored by the Kavli Foundation, invited leading neuroscientists and representatives from ten DOE National Laboratories to consider possible National Laboratory contributions to the BRAIN Initiative. "A consensus emerged that the DOE National Labs could make a distinctive and valuable contribution to the BRAIN initiative, and that this engagement would generate significant spin-offs impacting each of the DOE mission areas of Energy, Security, Science, and Environment."

In October 2015, a joint DOE-NIH workshop served to define the preliminary scope of DOE participation. In March 2016, the White House Office of Science and Technology Policy announced: *"Department of Energy (DOE): The DOE plans to invest $9 million to the BRAIN Initiative focused on the development of enabling technologies through access to the Office of Science User Facilities, with respect to three major themes: developing the specialized, high-resolution tools for measuring key neurological processes, developing the capabilities for obtaining a dynamic, real-time read-out of these measurements, and developing the integrated computational framework for analyzing and interpreting this dynamic multi-modal data. Developing the tools to integrate and synthesize multi-modal data on the brain and nervous system would be unprecedented and would inform other analyses of complex systems. A workshop will be held in FY 2016 to inform the priority requirements for developing novel biosensors and probes that can measure key molecular components or processes relevant to neuroscience."*

This Neuro-workshop was convened on June 2, 2016 at LBNL with participants from two National Laboratories (LBNL and PNNL), two BER/DOE User Facilities (EMSL and JGI) and JBEI to discuss capabilities and collaborative opportunities. The goals of the workshop were to:

1. Overview the unique and complementary capabilities and expertise of the participating institutes that are relevant to the BRAIN Initiative and to the DOE-stated contributions for the BRAIN,
2. Share the scientific visions for brain research at the participating institutes, and
3. Identify opportunities for inter-institute cooperation.

The workshop also discussed opportunities for applying neural technologies to advance the DOE mission in environment and energy, and benefits of technological synergies between the BRAIN and other national biomedical initiatives.

Background Internet Links: BRAIN Initiative at NIH; DOE in BRAIN; National Microbiome Initiative (NMI); The Challenge of Connecting the Dots in the B.R.A.I.N.

Opportunities for Multi-institute Collaborations

Introductory overviews of the brain-relevant capabilities at the participating institutes (pages 8-9) and their visions for brain science (pages 10-13) laid the foundation for open discussions on the DOE development of neuro-technologies in the context of the workshop goals (page 5). There was substantial interest among the participants to launch collaborative projects to develop advanced neuro-technologies to (a) link the activities of individual neurons and small populations of neurons to large-scale patterns of neural activity underlying sensations and behaviors in living animals, and (b) understand mechanisms of susceptibility and resilience to environmental exposures, role of metazoan neural function in soil and plant ecosystems, and diversity of risks for neurological diseases.

Three cross-cutting technological challenges were highlighted:

1. *In vivo* multi-modal and multi-scale measurement and manipulation of dynamic brain function in behaving individuals -- Develop biosensors and tools for electrical, chemical, and molecular interrogations of brain sub-regions, cerebral spinal fluid, blood-brain barrier, and for mapping the communication among behavior-determining neural networks.
2. High-resolution molecular and structural analyses of brain tissue sub-regions -- Develop small sample (e.g. single cell) capabilities for deep multi-omic profiling and micro-anatomy; develop rodent genetic models enabled by transgenic and genome editing methods to facilitate single-cell studies of neuronal subtypes.
3. Multi-modal data collection and analysis pipelines for data integration and prediction -- Develop tools and pipelines for multi-modal measurements that use common formats and analysis methods for interpretable and predictive results at scale.

There was broad agreement that the development of neuro-technologies would be accelerated by 'in-house' evaluations in complementary rodent and insect systems by closing the 'design-test-learn' loop.

Neuro-technological developments at DOE National Laboratories and User Facilities hold the promise to advance basic knowledge in environment and bioenergy that could be translated to applications relevant to other DOE-Science initiatives. Two broad directions were highlighted:

1. To promote sustainable ecosystems through multi-scale understanding of nervous systems and behaviors of soil metazoans, flying insects, and small animals in ecosystems.
2. To apply neuro-technologies to reveal mechanisms of individual diversity in structure and function of nervous systems, and how they impact organismal behavior, resilience, and survival in response to environmental stressors, microbiomes, and biosynthetic byproducts.

There was consensus that the participating institutes have sufficient existing capabilities to launch several synergistic collaborations towards these goals. As one example, understanding the cortical dynamics underlying individual variations in behavior and changes following environmental exposures could be accomplished by integrating the following complementary capabilities of the participating institutes. To enhance real-time causal perturbations of brain function, the DNA synthesis capabilities of the JGI would generate constructs for enhanced light-activated ion channels/pumps (optogenetic actuators) that would be optimized utilizing the synthetic biology tools and robotic pipelines of JBEI. Such bioactuators would be genetically targeted to subpopulations in the brains of flies and rodents (LBNL/JGI) to obtain cell-type specificity. To gain insight into neuronal dynamics, animals would be tested for specified behaviors while undergoing electrophysiologically recording combined with simultaneous modulation of neuronal activity with bioactuators (LBNL). Comparative analyses of metabolomics and proteomic profiles of cerebral spinal fluid and blood in live animals would interrogate the integrity of the blood brain barrier during behaving and after environmental exposures (LBNL). Excision of brain tissue would permit isolation of cell types for multi-omics analyses and investigations at

the molecular level (EMSL). Multi-omics data visualization tools would be used to integrate transcriptomics, proteomics, metabolomics (JBEI/EMSL) to generate metabolic hypotheses that could be interrogated by tissue imaging (LBNL/EMSL) and with genetically engineered rodents (JGI). Finally, extraction of interpretable and predictive features from large, high-dimensional time-series data sets would be enhanced by state-of-the-art statistical data analysis methods utilizing high-performance computing (LBNL). Together, these synergistic activities would be designed to provide an unprecedented comprehensive multi-scale understanding dynamic brain function in health, disease, and under environmental stress.

Follow up activities to develop inter-institute collaborations - *1 year tasks*

1. Visits to PNNL/EMSL and follow-up meetings to understand the existing capabilities and to identify complementary capabilities and immediate collaborative opportunities.

2. Develop pilot projects between LBNL and PNNL and understand the logistic challenges of large –scale scientific endeavors among institutes (see example above).

3. Explore existing JGI/EMSL co-proposal mechanisms.

4. Launch BRAIN-seminar series in Fall 2016 with web links to PNNL, JGI, JBEI, EMSL.

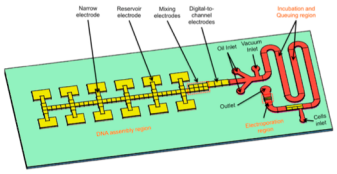
5. Communicate the contents of this report to stakeholders (lab management, DOE program managers, NIH program managers, philanthropic organizations) through dissemination of report, teleconferences and in person meetings.

Institutional Capabilities relevant to the BRAIN Initiative

Capabilities at EMSL Relevant to the BRAIN Initiative *– Harvey Bolton*

EMSL, a DOE Office of Science User Facility at PNNL, has developed an extensive tool kit of diverse technologies that are relevant to the study of the brain across broad spatial and temporal scales. The Cell Isolation and Systems Analysis (CISA) enables the isolation of individual cells or cell types for multi-omics analyses and biophysical investigations. Quantitative live cell fluorescence imaging is available to investigate individual cells and their relationships in tissue at single-molecule sensitivity. Molecular profiling is performed by transcriptomics using next-generation sequencing (5500XL SOLiD Sequencers) and ultra-sensitive proteomics (Mass Spectrometer: Ion Mobility Spectrometry, Time of Flight). Electron Microscopy provides ultrastructural information of neuronal cells at micro- to nano-scales. Various microscopy instruments enable multi-modal imaging with complementary chemical, structural and elemental information, and can be used for *in-situ* dynamic imaging in native tissue environments at high spatial resolution. Advanced Mass Spectrometry (MS) enables global proteomic, metabolomic, and glycomic analyses and MS-based imaging of biological systems and cells. A new 21 Tesla Fourier transform ion cyclotron resonance mass spectrometer provides high-mass resolving power and mass-accuracy measurements of biomolecular and organic compounds, including intact proteins, splice variants and various proteoforms. Nuclear Magnetic Resonance (NMR) and Electron Paramagnetic Resonance (EPR) provide high-resolution protein structure information, analysis of metal-centers and redox chemistry of proteins. EMSL technological capabilities are strengthened by molecular science computing, high-performance computing, sophisticated data analytics and data integration procedures. EMSL capabilities have been applied to PNNL neuroscience projects to generate a voxelated 3-D map of the brain proteome to characterization of Lewy Bodies in Parkinson's disease and a developmental map of nicotine receptors in the brains of juvenile rats.

Capabilities at JBEI Relevant to the BRAIN Initiative *- Blake Simons, Hector Garcia Martin*



Microfluidics chips for DNA assembly into a plasmid and subsequent eletroporation into a cell.

JBEI at LBNL is devoted to producing the scientific and technological basis to enable lignocellulosic biofuels. Several of the technologies created in this endeavor are widely applicable and can help advance the DOE support of the goals of BRAIN initiative. For example, the microfluidics capabilities at JBEI have mainly been used to automate synthetic biology, but could, in principle, be used for automation of brain cell culturing and phenotyping (e.g. through multi-omics analyses). Furthermore, the capabilities for metabolic flux analysis and predictions could be used to gain a clearer understanding of neural metabolism. This is particularly relevant since a variety of neurological dysfunctions have been related to metabolic dysfunctions. Multi-omics data visualization tools such as Arrowland can be used to intuitively integrate transcriptomics, proteomics, metabolomics and fluxomics data. High-throughput targeted peptide and proteomics analysis workflows developed at JBEI can be used to produce quantitatively accurate proteomic information for brain cultures. Synthetic biology tools and robotic pipelines, such as j5 and DIVA, can aid in developing biosensors for molecular interrogation of brain subregions. Finally, tools and standards have been developed at JBEI to manage, collect and visualize all data and metadata being created in this effort (the Experiment Data Depot). These tools and standards could be applied to facilitate the data-intensive methods to be used in the BRAIN initiative and provide a single repository of experimental data for all national labs involved, which would help create a cohesive initiative.

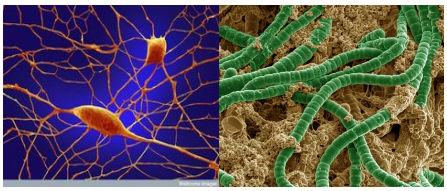
Capabilities at the JGI Relevant to the BRAIN Initiative *– Axel Visel*

The JGI is a Department of Energy User Facility at LBNL that provides a wide spectrum of genomic, functional genomic, and computational capabilities to users working on DOE mission-relevant questions related to energy and the environment. Many of these capabilities could potentially also be leveraged to support the goals of the BRAIN Initiative. Specific capabilities include: (1) Next-Generation Sequencing. The JGI has a several Illumina and Pacific Biosciences sequencers that can be used for the cost-efficient generation of large amounts of sequence. In the context of the BRAIN initiative, this capability could be used to perform transcriptomic and epigenomic studies on brain tissues. (2) DNA Synthesis. The JGI has a DNA synthesis platform that can be used to generate large (tens of thousands of basepairs) synthetic DNA constructs. For example, this capability could be used to generate DNA constructs for genetic interrogation of brain function in mice and other model systems. (3) Single-cell technology. The JGI has a Fluidigm F1 instrument and is currently establishing single-cell Drop-Seq technology. Both methods can in principle be used to perform single-cell transcriptomic studies of brain tissues. (4) Computational capabilities. The JGI has extensive experience in computational processing of DNA sequence data. This includes highly trained bioinformaticists, a comprehensive set of in-house developed computational tools and resources, and computer platforms that include access to High-Performance Computers (HPC) at NERSC. These capabilities could be leveraged in the computational analysis of BRAIN-related sequence data sets. (5) Microbial community sequencing studies. In the context of the BRAIN initiative, these capabilities are relevant to assessing interactions between microbiomes and brain function in changing environments or after exposure to bio-energy by-products. A full list of JGI sequencing products is available at http://jgi.doe.gov/user-program-info/product-offerings/ .

Capabilities at LBNL Relevant to the BRAIN Initiative *-- Kris Bouchard*

Interdisciplinary teams of LBNL biologists, computer scientists, chemists, materials scientists, physicists, and engineers from Biosciences, Computing, Energy Sciences and Physical Sciences areas are engaged in several projects to design novel neuro-technologies to enable multi-scale measurement and manipulation of neuronal activity from awake behaving animals. These technologies are to be integrated with molecular and micro-anatomical analyses of cells in circuits and will be available to support the DOE role in BRAIN, environmental and energy missions, and other national initiatives. Specific capabilities include: (1) Design and post-processing of electronic interfaces and data acquisition hardware to enable scalable, massive channel count, multi-scale *in vivo* electrical measurements and closed-loop manipulations of neuronal activity; (2) Novel methods for getting light to deep-brain structures through the utilization of up-converting nanoparticles for measurement and manipulation of neuronal activity; (3) State-of-the-art statistical data analyses methods for extraction of interpretable and predictive features from high-dimensional data sets; (4) Computational resources and frameworks for the neuroscience community. [Uberon: A multi-species anatomy ontology and knowledge base; CRCNS: Collaborative Research in Computational Neuroscience, a data repository for neurophysiology data]; (5) Non-invasive human brain imaging [1.5 T MRI, PET, Biomedical Isotope Facility]; (6) Multiple genetic tools with differential spatiotemporal expression in the brains of flies and mice that can be used for targeting biosensors and bio-actuators to neuronal subpopulations; (7) Live single-cell imaging and extraction and multi-omic analyses of sub-cellular samples from tissue slices and brain regions; (8) Multiple model systems (e.g. flies, mice, rats) for testing and utilization of developed neuro-technologies in awake behaving animals in response to environmental challenges, by-products of biofuel production, and changes in gastric microbiomes. Items 1 – 4 make extensive use of NERSC and the Molecular Foundry, while 5-8 leverage expertise and equipment in the Bioscience Area [e.g. Rodent Behavioral Core, Bioimaging Center].

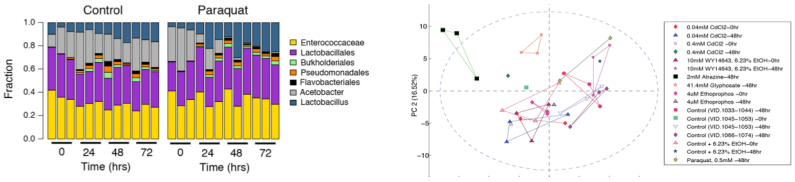
Brain Science Visions at the Participating Institutes

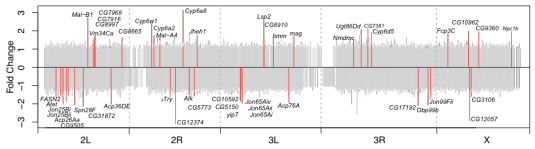


Janet Jansson PNNL
Microbiome "Brain" storming

Microbiomes are complex systems that have interactions and signaling that occur between hundreds to thousands of cell types and modeling of these communication networks can serve as an analogy to the brain.

- PNNL has made major investments ($25 million) in microbiome research; including understanding and prediction of the impacts of perturbations on human and environmental microbiomes and integrative assessment of plant-soil-microbiome interactions.
- A direct connection between microbiomes and brain research is via the gut-brain axis; PNNL is studying the impact of different external factors on behavior in humans and in mouse models via gut microbiome to brain signaling. These studies take advantage of multi-omics and imaging capabilities at PNNL and EMSL.
- There is a need for advances in computation, multi-omics and imaging to be able to decipher and understand the roles that specific microbes and metabolites play in gut-brain interactions.



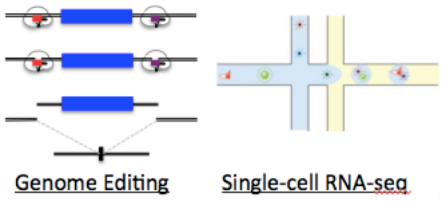


Susan Celniker, LBNL
Genomic Responses to Environment

Exposure-induced changes in the Microbiome, Host Transcriptome and Metabolome

In Drosophila, we can build a powerful system for rapidly identifying the molecular basis of neurological phenotypes – an important step on the road to understanding the brain.
Key areas we are developing:

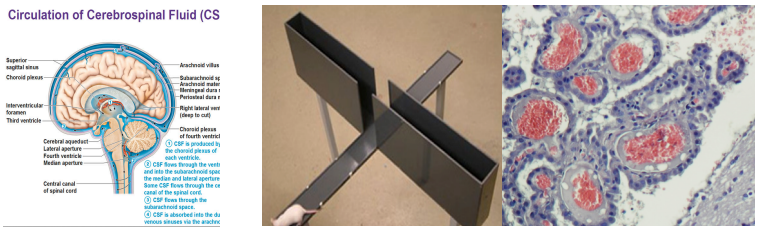
- Behavior as a readout for complex perturbations due to environmental exposures,
- Mapping genome functions in neurons,
- Tools for studying the interplay between gene expression and neural activity in *Drosophila melanogaster*, and
- Mapping molecular interactions in any cell accessible by targeted expression (protein-protein, protein-RNA, protein-DNA, RNA-DNA).



Diane Dickel and Axel Visel, LBNL
Cutting-Edge Genome Engineering and Single Cell Transcriptomics in the Mouse

The Mammalian Functional Genomics Program at LBNL has established cutting-edge, high-throughput *in vivo* mouse genome engineering and transgenic capabilities and large-scale enhancer resources.

- In <3 years, we have generated >60 engineered mouse lines and could easily make a variety of mouse models for use as part of a DOE-led BRAIN Initiative project.
- Our group is implementing and developing single-cell transcriptomics (RNA-seq) technologies that can be used on *ex vivo* mouse tissue, for example to profile neuronal cell populations.
- These capabilities can be paired with our mouse transgenic pipeline, along with our VISTA enhancer resource and expertise of gene regulation, to generate new insights into brain function.
- These methods are not limited to mammals and can be broadly applied to many eukaryotic organisms.

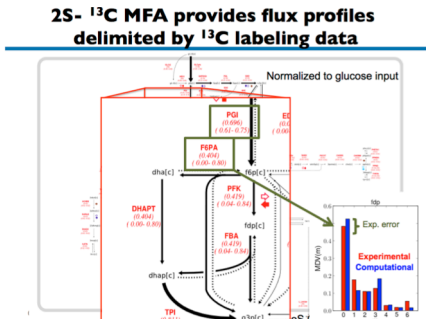


Andrew J. Wyrobek, LBNL
Individual Variations in Behavioral Responses and Neurological Risks

Our goal is to understand the CNS mechanisms by which environmental stressors to the young brain increase risks for late-onset neurological deficits such as memory loss, anxiety, CNS vascular abnormalities, amyloid plaque formation and tissue pathologies associated with Alzheimer and neurological diseases. Our findings to date are based on molecular and cellular CNS tissue damage and behavioral responses in outbred rats exposed to low LET (low linear energy transfer beams, e.g., x-rays in radiotherapy) and high LET (e.g., space radiation) forms of ionizing radiation.

- Multi-omic profiling of brain regions has generated hypotheses of CNS pathologies and predicted vascular abnormalities and amyloid plaques have been confirmed by immunohistochemistry.
- The choroid plexus, which produces cerebral spinal fluid, is sensitive to radiation damage that can persistent for the lifetime of the animal.
- Differential molecular profiling of brain regions in animals with variations in behavior responses to exposure has yielded hypotheses for the mechanisms of susceptibility and resistance.

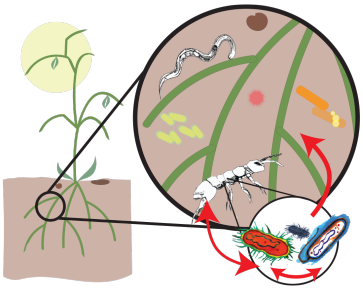
Our vision is to apply advanced neuro-technologies to understand the molecular mechanisms of individual variation in memory deficits and anxiety, and to predict individual risks for neurological deficits and diseases. Underlying questions are: (1) what is the relationship between molecular expression and electrophysiology of neural circuits in animals that are sensitive versus resistant to environmental stressors, and (2) can reliable CSF and blood biosensors be developed to predict individual variations in susceptibility.



Hector Garcia, LBNL
Fluxes as Predictors of Function

Brain energy metabolism is central to all cellular processes that maintain neuronal functionality.

- The most recent work on neural metabolism uses simplified stoichiometric models and flux techniques that rely on strong assumptions (Flux Balance Analysis, FBA).
- At JBEI, we have developed techniques to combine ^{13}C labeling data and genome-scale models for measuring and predicting metabolic fluxes for *E. coli* and *S. cerevisiae*. ^{13}C-labeling eliminates the need for unnecessary assumptions and genome-scale models provide comprehensive description of metabolism. These techniques could be used for neurons and other brain cells.
- We are also developing new software tools to facilitate flux analysis and multi–omics data visualization.

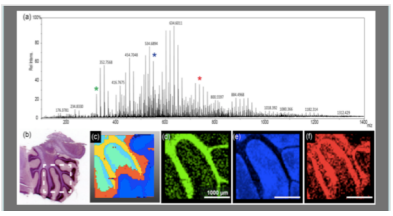


Javier A. Ceja-Navarro (LBNL)
Insects as Mediators of Ecosystem Service

We have defined important associations among arthropod systems and their microbiomes and we are investigating the contribution of the gastric microbiota in lignocellulose transformation processes and detoxification. Our studies show that microbiome of soil fauna is a compartment of soil nutrient cycling.

- Screening the microbiome of soil metazoans demonstrated that each metazoan evaluated contained a defined microbiome that varied with habitat and diet.
- By manipulating metazoan microbiomes, we successfully removed traits from metazoans and associated these changes in traits to modifications in host's fitness.
- Our research is based on several soil metazoan model systems. For example, the passalid beetle is a model for a carbon cycling machine. And the coffee berry borer, the most important coffee pest, survives toxic levels of caffeine in coffee beans.

Our findings suggest that environmental changes in microbiomes of soil metazoans can alter both behavior and fitness. This suggests that microbiome-induced behavioral changes in soil fauna may be mediated by environmentally-induced changes to neural functions.



Steven Wiley, EMSL
Approach to Neurobiology at EMSL

Research in neurobiology at EMSL has primarily focused on developing approaches to analyze the distribution and identity of bioactive molecules in intact tissue and biological samples. These approaches include:

- Spatial mapping of bioactive molecules
  - Primarily mass spectrometry-based imaging, such as nanoDESI, MALDI, SIMS, nanoSIMS
  - Multiple imaging modalities can be combined to yield unique insights
- Compositional analysis of complex biological samples
  - Proteomics of fluid and tissue samples using multiple, ultra-sensitive approaches
  - Transcriptomics analysis of very small samples
  - Can use both top-down and bottom-up proteomics and combine with transcriptomics
- Combination of selective isolation and compositional analysis
  - Flow cytometry
  - Laser-capture microdissection
- Automated data collection and integration using statistical and mechanistic models

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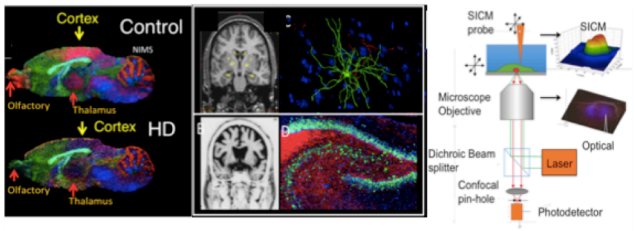

Cynthia McMurray, LBNL
Technology and biology: Predicting life and death responses of neurons

How brain cells respond to environmental exposures and the nature of the molecular signals that determine cell survival or cell death are poorly understood. Neural cells do not operate in isolation, and brain tissue comprises heterogeneous networks of neurons and supporting cells. Our research goals are to understand signaling mechanisms in functional neural circuits and how they are affected by environmental exposures, and in age-related neurodegenerative diseases. We conduct our research using mouse models.

- Apply integrated biology approaches to build novel imaging tools to link the spatial position of neurons to their function without disrupting their *in situ* neuronal circuitry. This includes multi-scale imaging, single-cell capture from tissue and characterization by high-throughput multi-omics.
- Identify affected neural circuits by mass spectrometry imaging, define their electrical signaling properties, characterize their gene expression and metabolic profiles in order to understand the mechanisms that predict cell fate in brain tissue exposure to environmental challenges and in brain tissue of individuals with age-related neurodegenerative diseases.

Workshop Participants

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***Institute POCs and alternates for writing this report.**