



2012 NESM Spring Symposium @ The Marine Biological Laboratory, Woods Hole

FRIDAY, MAY 4, 2012

Symposium Schedule:

- 9:00 AM Registration (Swope Center):** Coffee and Refreshments
- 10:00 AM Welcome (Meigs Room):** Louie Kerr, *NESM Biological Sciences Director*
- 10:10 AM “Confocal and super-resolution imaging of muscle”,** Elizabeth Brainerd, Ph.D., *Brown University*
- 10:50 AM “Microscopy and authenticity in the art museum: How microscopes shed light on the origins of cultural artifacts”,** Richard Newman, *Museum of Fine Arts, Boston*
- 11:30 AM Vendor/Poster Session**
- 12:30 PM Lunch (Swope Center)**
- 1:15 PM Keynote: “TEM: The key tool for nanotechnology”,** Barry Carter, D.Phil., Sc.D., *University of Connecticut*
- 2:15 PM Afternoon Break:** Coffee and Refreshments
- 3:00 PM “Ion microprobe analyses at WHOI: Using micron-scale measurements to understand global scale processes”,** Brian Monteleone, Ph.D., *Woods Hole Oceanographic Institution*
- 3:40 PM “The juvenile ALS2 gene product Alsin encodes a protein that regulates IGF-1 receptor endocytosis and cell signaling”,** Justin Topp, Ph.D., *Gordon College*
- 4:20 PM “Making improved neural activity indicators: Genetics and calcium imaging methods”,** Trevor Wardill, Ph.D., *Marine Biological Laboratory, Woods Hole*
- 5:00 PM Closing Remarks:** NESM Board

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CORPORATE VENDOR EXHIBIT**

We extend our deepest gratitude and thanks to all our exhibiting vendors for their financial support. A portion of the cost for a table display is used to reduce registration costs for students and retirees.

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ABSTRACTS & BIOS

10:10 am – **Confocal and super-resolution imaging of muscle**

Elizabeth Brainerd, Ph.D., *Brown University*

Abstract:

Striated muscle is a structurally beautiful and dynamic material that powers animal movement. Since the middle of the 20th century, the sliding filament model of muscle contraction has been a valuable tool for explaining muscle contraction at the molecular level. Recent proposed modifications to the sliding filament model, such as the titin winding filament hypothesis, offer more complete explanations of muscle behavior, but have been hard to test. We are using immunohistochemistry, confocal microscopy and super-resolution microscopy to visualize myofilaments in relaxed and contracted muscle. We have not yet succeeded in testing these new models, but results from our preliminary microscopy studies of zebrafish larvae *in vivo* and rabbit psoas muscle fibers *in vitro* look promising.

Bio:

Professor Elizabeth Brainerd received her doctorate from Harvard University in 1991, where she studied the functional morphology of air-breathing fishes with Professor Karel Liem. For her post-doctoral work, Dr. Brainerd studied biomechanics with Professor Tom McMahon in the Division of Applied Sciences at Harvard, supported through a Junior Fellowship from the Harvard Society of Fellows. From 1994-2005, Dr. Brainerd served as Assistant then Associate Professor at the University of Massachusetts Amherst, and then Professor at Brown University starting in 2006. Her research interests center on musculoskeletal biomechanics in vertebrate animals, with particular emphasis on developing and using advanced imaging technologies to study live animals in motion. She directs the XROMM development group (www.xromm.org) and is a member of the QSTORM collaboration (www.qstorm.org). Professor Brainerd received a Lilly Teaching Award from the University of Massachusetts, a CAREER award from the National Science Foundation and is a Fellow of the American Association for the Advancement of Science.

10:50 am – **Microscopy and authenticity in the art museum: How microscopes shed light on the origins of cultural artifacts**

Richard Newman, *Museum of Fine Arts, Boston*

Abstract:

Art forgers and forgeries have long been a source of fascination to laymen and scholars alike. This talk begins with two of the most famous art forgers of the 20th century, then discusses some objects in the collection of the Museum of Fine Arts whose authenticity is questionable. Many scientific methods are now used to address questions of authenticity, but an SEM with microanalysis capability remains one of the most versatile.

Bio:

Richard Newman is Head of Scientific Research at the Museum of Fine Arts, Boston, where he has worked since 1986. Prior to this, he worked as both an objects conservator and scientist at Harvard University's Fogg Art Museum. He oversees all scientific research related to the museum's collections, and frequently collaborates with researchers from other museums, including, in recent years, a project on Inkan qero cups. He has received fellowships for research in India, Spain and Japan, and was coauthor of *Examining Velazquez*, which received the 1991 Award for Distinction in Scholarship and Conservation, a joint award from the College Art Association and National Institute for Conservation.

1:15 pm – **TEM: The key tool for nanotechnology**

Barry Carter, D.Phil., Sc.D., *University of Connecticut*

Abstract:

The transmission electron microscope is now about 80 years old. I have now been using the TEM for 40 years! When I started, the TEM was a slide rule (I missed the earliest stages, clearly); now it's a PC—and not always a Mac! It's actually becoming more powerful and more of a black box, which should be a real concern to all of us. I will discuss some of the exciting possibilities for the future using examples from today and from those recent 40 years, illustrating why we need to emphasize teaching and training, but keep remembering history and reading papers that appeared before the age of electronic journals! I'll emphasize that we should, of course, always use the microscope that fits the task, rather than working the other way round. I'll overdo the pedagogy, but hopefully also advance my opinion that our two cultures in microscopy can intercept and interact beneficially, even if they don't very often. The topics I'll illustrate will include nanoparticles, interfaces, local chemistry, but always, microscopy.

Bio:

Barry Carter is a Professor at UConn and Head of the Department of Chemical, Materials & Biomolecular Engineering. Before moving to UConn in 2007, he spent 16 years as the 3M Harry Heltzer Multidisciplinary Endowed Chair in the Department of Chemical Engineering & Materials Science at the University of Minnesota. Prior to that, he spent 14 years in the Department of Materials Science & Engineering at Cornell University leaving there as a full

Professor. Barry earned his bachelor's degree at Cambridge University, his master's at Imperial College and his doctorate at Oxford University, then another doctorate at Cambridge. He has published around 300 scientific journal papers, more than 400 conference papers, and co-authored 2 textbooks. He was honored with a Guggenheim Fellowship in 1985 and a von Humboldt Senior Award in 1997. He was the Bernd Matthias Scholar at Los Alamos National Lab in 1997-98, the 2004 Jubilee Professor at Chalmers University in Göthenburg and a Visiting Fellow at Peterhouse in Cambridge during 2005. He is a Fellow of AAAS, MRS, MAS, ACerS and RMS. He is the Editor-in-Chief of the Journal of Materials Science (over 26,000 cites in 2010), which has more than doubled its IF since he took this position in 2004. He chairs the MRS Awards Committee, was the 1997 President of MSA, and is the current President (2011- 14) of the International Federation of Societies for Microscopy. In his research, he has used TEM, SEM, AFM, STM and VLM to study metals, semiconductors, polymers, ceramics and even some biomaterials. His favorite material is aluminum oxide – it's a gem of a material.

3:00 pm – Ion microprobe analyses at WHOI: Using micron-scale measurements to understand global scale processes

Brian Monteleone, Ph.D., *Woods Hole Oceanographic Institution*

Abstract:

The Northeast National Ion Microprobe Facility (NENIMF) at WHOI is an open laboratory that facilitates micron-scale chemical analyses through focused ion beam sputtering and secondary ionization mass spectrometry (SIMS). The facility houses a Cameca 1280 Ion Microprobe (installed in 2005), ideal for mass resolution up to ~7000, and a lower mass resolution Cameca 3f (installed in 1978), which is currently undergoing major renovation to computer-based Labview operation. The lab focuses on a variety of Earth and Ocean Science projects including (but not limited to) volatile (CO₂, H₂O, F, S, Cl) concentrations in melt inclusions and apatites from various volcanic/tectonic settings, chemical isotopic compositions (e.g. B, S, O) in various mineral phases from rocks from different tectonic settings, and measurement of paleo-proxies for seawater temperature and pH in corals and microfossils. This talk will give an overview of the facility operation and recent scientific highlights.

Bio:

Brian Monteleone completed his MS in Geosciences at The University of Arizona in 2000 and his Ph.D. in Earth Sciences at Syracuse University in 2007. His work focused on combining Noble Gas thermochronology (40Ar/39Ar dating) with ion microprobe U/Pb geochronology and trace-Rare Earth Element chemistry to constrain the exhumation of ultra-high pressure rocks in Papua New Guinea. His post-doctoral work at Arizona State University focused on ion microprobe U and Th measurements and laser ablation He measurements to develop an *in situ* (U-Th)/He dating method for apatite and zircon. He joined NENIMF in March, 2011 as a research associate and analyst.

3:40 pm – **The juvenile ALS2 gene product Alsin encodes a protein that regulates IGF-1 receptor endocytosis and cell signaling**

Justin Topp, Ph.D., *Gordon College*

Abstract:

ALS is a heterogeneous group of disorders that result in motor neuron death and are usually fatal. ALS2 is a rare juvenile form of ALS that is caused by recessive loss-of-function mutations in the gene encoding Alsin. We and others have shown that Alsin is a large protein with multiple domains involved in GTPase regulation. In the current study, we show that Alsin functions in IGF-1 receptor trafficking. Overexpression of a putative dominant-negative version of Alsin inhibits IGF-1 receptor endocytosis and blocks IGF-1 mediated signaling as determined by immunofluorescence and biochemical assays. Furthermore, loss of IGF-1 signaling with dominant-negative Alsin overexpression decreases cell survival, which was also observed in cells depleted for Alsin. Taken together, these data reveal that Alsin regulates IGF-1 mediated pathways and suggests a potential novel mechanism for ALS disease progression.

Bio:

Dr. Justin Topp is Associate Professor of Biology at Gordon College in Wenham, MA. He received the B.S.E. in Biomedical Engineering from the University of Iowa in 2000 and Ph.D. in Biological Chemistry from the University of Texas-Southwestern Medical Center at Dallas in 2005. After graduate school, he pursued post-doctoral studies in Molecular Biology and Biochemistry at the same institution before becoming Assistant Professor of Biology at North Park University in Chicago, IL in 2008. Dr. Topp has a broad and diverse research background, having worked in the areas of membrane trafficking, cell signaling, alternative splicing, and molecular microbiology. His current work is focused on two areas: 1) using biotechnology techniques to track tick-borne infectious agents, and 2) using biochemical and immunofluorescence studies to characterize the ALS disease protein, Alsin.

4:20 pm – **Making improved neural activity indicators: Genetics and calcium imaging methods**

Trevor Wardill, Ph.D., *Marine Biological Laboratory, Woods Hole*

Abstract:

Imaging-based detection of calcium flux in neurons using engineered fluorescent protein scaffolds has enabled improved detection of neuronal activity. Genetically-Encoded Calcium Indicators (GECIs) allow for relatively non-invasive activity sensing, where hundreds of neurons can be monitored simultaneously. To produce GECIs with improved performance, we developed a neuron-based screening pipeline to test thousands of GECI variants. Variants of the Green Fluorescent Protein/calmodulin-based calcium sensor, GCaMP3, were made by site directed mutagenesis. GCaMP3 variants were initially tested in cultured rat primary hippocampal neurons transduced with lentiviruses using a custom built high throughput automated field stimulation and EMCCD fluorescence imaging platform.

Improved GCaMP3 variants identified by the *in vitro* screen were characterized *in vivo* in cortical neurons in mice, mechanosensory neurons in *C. elegans*, in the *Drosophila* larval neuromuscular junction (NMJ) and in projection neurons (PN) of the adult fly antennal lobe. For the NMJ preparation, we used a suction pipette to directly stimulate single action potentials in motor neurons with 300 μ s electrical pulses while imaging calcium responses in their presynaptic boutons using an EMCCD camera. The best of these GCaMP3 variants are then tested in PNs responding to odor stimuli. By targeting GECIs to specific PNs and glomeruli, and using different odor concentrations, we are able to investigate GECI performance under a variety of activity regimes. GECI technology will aid in the dissection of neuronal circuits by allowing for correlation of specific neuronal activity with behaviors *in vivo*.

Bio:

Dr. Trevor Wardill considers himself a neuroscientist with expertise in electrophysiology and functional imaging (2-photon and EMCCD). His research interests have progressed from molecular biology/population genetics to intracellular and extracellular neurophysiology in *Drosophila*. Dr. Wardill has used transmission electron microscopy to help develop transgenic *Drosophila* with pure ultraviolet vision and wild-type structure to allow the dissection of color and motion visual circuits. To further understand the importance of color and motion visual circuits, he helped developed behavioral assays of *Drosophila*. Drawing on these experiences, Dr. Wardill's post at Howard Hughes Medical Institute allowed him to further improve his imaging skills using genetically induced and chemically introduced calcium indicators for dissociated cells and *in vivo* experiments. For analysis of such data, Dr. Wardill has written and refined numerous programming routines within Matlab to allow automated image-based segmentation and data analysis. Now in the Hanlon Lab, he has elucidated a range of stimulation protocols necessary to stimulate *Doryteuthis pealeii* skin iridophores and he performs spectral measurements of their reflectance changes. Furthermore, by adapting a protocol developed by Gonzalez-Bellido, Dr. Wardill has imaged whole mount sections of thick cephalopod skin to allow production of 3D reconstructions of the neural network that innervates the skin. Aside from pure science, he possesses the hands-on talent to build custom electrophysiology and imaging rigs for a variety of specialized *in vivo* experimentation.

29TH ANNUAL SPRING SYMPOSIUM

POSTER EXHIBIT

Spatial distribution of human gut bacteria in gnotobiotic mouse models

Y. Hasegawa, *Brown University and Marine Biological Laboratory*

Activities of intestinal bacterial communities are crucial for human health. The goal of this project was to develop a novel imaging-based assay to study spatial arrangements of bacteria in the intestines of gnotobiotic mouse models. Gnotobiotic mouse models used in this study were created by inoculating up to fifteen human intestinal bacteria into germ-free mice. In order to develop an imaging assay, we (1) explored protocols to preserve spatial arrangements of luminal contents in intestinal sections, (2) demonstrated the ability to simultaneously distinguish multiple bacterial species using fluorescence in situ hybridization (FISH) combined with spectral imaging and (3) explored image analysis methods to capture bacterial spatial distribution patterns in intestinal sections. By using a spectral imaging protocol developed in this study, multiple bacterial species labeled with FISH probes were simultaneously distinguished in their native environmental conditions. Our analyses of bacterial spatial distribution revealed enrichment patterns of bacterial cells in certain areas of the intestinal sections at multiple different scales.

Optics Course Poster: Building a color microscope

P. Hesketh, D. von Maluski, D. Melzar, *UMass Amherst*

We have designed and built a transmitted light microscope from optics components in our Optics course at the University of Massachusetts Amherst. We have also added a filter set in front of the camera to enable viewing in red, green, and blue separately. Using ImageJ, we can recombine the three color images to create a full-color image.

Optics Course Poster: Building an epi-fluorescence microscope

C. Brueckner, V. Porter, A. Gillooly, *UMass Amherst*

We have designed and built a transmitted light microscope from optics components in our Optics course at the University of Massachusetts Amherst. We have also added an epi-fluorescence component in order to visualize red fluorophores.

Optics Course Poster: Building a confocal microscope

H. Kincaid, Z. Bodine, A. Barnes, *UMass Amherst*

We have designed and built a transmitted light microscope from optics components in our Optics course at the University of Massachusetts Amherst. We have researched the modifications needed to change our system into a scanning confocal fluorescence system.

