Welcome to BEMF News

BEMF News will be brought to you quarterly from the Pacific Biomedical Research Center Biological Electron Microscopy Facility at the University of Hawai‘i at Mānoa.

Each quarter we will highlight upcoming events in the facility and the happenings within the microscopy community at large.

We will also keep you informed of the interesting research that takes place in our lab in our Featured Research section. Here we will profile the researchers, their interests and latest discoveries.

Please contact us if you have news or suggestions for future editions!

Introduction to PBRC’s BEMF

The Biological Electron Microscope Facility, or BEMF, was established in 1984 by the Pacific Biomedical Research Center (PBRC) here at the University of Hawai‘i at Mānoa under the direction of Dr. Richard D. Allen. BEMF has grown steadily since its inception in its instrumentation, expertise, and use.

We are a multi-user shared resource core facility which provides biological/biomedical researchers with modern, state-of-the-art instrumentation, training, and services for conventional and energy-filtering transmission electron microscopy, field emission scanning electron microscopy, brightfield, DIC and epifluorescence light microscopy, and laser scanning confocal microscopy.

In just the past 5 years BEMF has served faculty and researchers from over 50 laboratories in Natural Sciences, Tropical Agriculture and Human Resources, Engineering, Medicine, Ocean and Earth Sciences and Technology, and PBRC, as well as visiting investigators from BRIN partner, mainland and foreign institutions.

The BEMF operates on a recharge fee system through the Research Corporation of the University of Hawai‘i, and is available to anyone within the State and beyond.

Skilled assistance and expertise are provided by Dr. Richard D. Allen, Director, Dr. Marilyn F. Dunlap, Manager, Tina M. (Weatherby) Carvalho, Facility Supervisor, and Tanya M. Michaud, Research Associate. Together, BEMF personnel comprise nearly 100 years experience in microscopy.
Instruments and Preparative Equipment

At BEMF our LEO 912 Energy Filtering Transmission Electron Microscope utilizes the latest technologies for conventional TEM imaging as well as selected energy filtering for elemental analysis. It is equipped with a cooled Proscan CCD camera and a powerful image analysis package.


For laser scanning confocal microscopy our Bio-Rad MRC 1024 is mounted on a Nikon Optiphot II upright microscope with software for 3D reconstruction and other presentation modes. Its Krypton/Argon laser can simultaneously excite three fluorophores.

We also have an Olympus BX51 upright compound microscope with widefield epifluorescence and an Olympus SZX12 stereo zoom microscope. Each can be fitted with our scientific grade digital camera. Mac and PC workstations are available for image analysis.

BEMF also houses a variety of preparative equipment including conventional and cryo-ultramicrotomes, a freeze-fracture device, critical point dryer, sputter coater, vacuum evaporator, and vacuum and microwave ovens.

Training

We offer one-on-one or group training in a number of different techniques and on all of our instruments.

Training on the SEM is fun and easy; the introductory session takes about three hours and, with another hour or so supervised session, users are familiar enough with the microscope to get excellent pictures. Preparation of biological specimens for SEM may be as easy as air drying and quick mounting and coating, or complex, requiring a full day of chemical fixation, critical point drying, and precise mounting and sputter coating.

The TEM is somewhat more complex to use, but the real challenge is in specimen preparation. We are able to train individuals in all aspects of specimen preparation for viewing of ultrathin sections or negatively stained particles, or we can perform any part as a service.

Specimen preparation for light, epifluorescence or laser scanning confocal microscopy takes many forms. Generally we can duplicate or modify many common techniques, including fixation, application of specific fluorescent dyes, and fluorescent immunolabeling.

We do not offer courses for academic credit at this time.

The BEMF is partially supported by a Biomedical Research Infrastructure Network award, RR-16467, from the National Center for Research Resources, National Institutes of Health.
BEMF Service

Our staff is available to perform any or all parts of specimen preparation and microscopy that you choose not to do yourself. This may include everything from fixing the sample to printing final pictures. Technician time is charged by the hour, as are the instruments, and we can provide the chemicals and supplies necessary for cost plus a small handling charge. This allows you to budget your time, money and expertise by choosing which parts of the microscopy you wish to do yourself vs. which parts for which you are willing to pay. We are always willing to discuss your project and give a general cost estimate without any obligation. Check with us, our brochure or web site for a fee schedule.

Upcoming Microscopy Conferences

Microscopy & Microanalysis 2003 will be held in San Antonio, Texas at the San Antonio Convention Center August 3-7, 2003.

The Pre-Meeting congress will feature the 2nd International Congress on Biophotonics August 2-3.

Special Meeting Topics will include Biological Applications of Optical Microscopy, Nanotechnology, and an extensive equipment exhibition. For full details, registration and submission procedures visit: http://www.msa.microscopy.com/MMHomePage.html

Microscopy & Microanalysis 2005 will be held here in Honolulu, Hawai‘i, July 31-August 4, 2005 at Hawai‘i’s Convention Center. With 3000 expected participants, this will be the 63rd Annual Meeting of the Microscopy Society of America and the 39th Annual Meeting of the Microbeam Analysis Society.

Other participating Pacific Rim societies include the Committee of Asia-Pacific Societies for Electron Microscopy and other southeast Asian societies (CAPSEM), the Australian Microscopy and Microanalysis Society (AMMS), Microscopy New Zealand (MNZ), and the International Metallurgical Society (IMS). Besides a diverse scientific program covering all types of microscopy, it includes an extensive exhibit of instrumentation and support equipment.

Microscopy Society of America
Hawai‘i Local Affiliate Society to form

The Microscopy Society of America was formed in 1942 primarily for electron microscopists. Over the years it has come to embrace all types of microscopy, microscopical techniques, and the various types of research and techniques associated with microscopy. The society has a formal journal, Microscopy and Microanalysis, and a large and well-attended annual meeting each August. Many areas of the United States have Local Affiliate Societies (LAS) which periodically have meetings, seminars, and workshops. These local societies foster exchange of ideas, techniques, and formation of collaborations. Our vision for a local microscopy society includes starting a seminar series and offering MSA-sponsored Mainland speakers, as well as organizing workshops. Depending on interest, we could hold quarterly, bi-annual or annual meetings, including informal get-togethers and/or more formal meetings with presentations and posters. If you are interested in joining or helping to organize a local society, please contact Tina at 808-956-6251 or tina@pbrc.hawaii.edu
We are elaborating the cellular role of a conserved gene, fli-I, which encodes a presumed actin-associated, regulatory factor. Such proteins play a major role during the rapid reorganization of the cytoskeleton, for example during neoplastic transformation, in motile cells, during cell division, and in growing axons during embryonic development. The ability of cells to reorganize their cytoskeleton determines the metastatic potential of cancer cells, and actin-associated proteins have been identified in degenerative diseases of the musculature.

Genomic analyses have identified homologues of the fli-I gene in Caenorhabditis, Drosophila, mouse, and humans. Defects in the gene cause either flightlessness or embryonic lethality in Drosophila, and early embryonic lethality in mice. Because the severity of the loss-of-function mutant phenotypes, fli-I is refractory to genetic analysis. An alternative strategy to glean information on the cellular role of a gene product is ectopic expression, in which gain-of-function effects are produced because the protein is expressed in excessive amounts or at inappropriate times and in tissues usually not transcribing the gene. Drosophila is particularly amenable to this approach, since the transcription of transgenes can be controlled by tissue-specific and developmentally regulated enhancers. Whereas general overexpression of regulatory proteins frequently causes death because of severe interference with fundamental developmental mechanisms, restricted expression in cells and organs that are dispensable for the survival of the organism is usually tolerated.

The support cells of mechanosensory bristles and hairs of Drosophila contain several bundles of cross-linked actin filaments, which serve to support this structure during the extension phase of the cells. After completion of this process, the chitinous cuticle rapidly hardens and replaces the mechanical function of the filament bundles, which leave grooves and ridges in the surface of the bristles. These ridges can be seen with a scanning electron microscope. Thus, the surface structure of the bristles, and the bristle shape itself are sensitive indicators for perturbances of actin filament organization.

Overexpression of fli-I in the mechanosensory bristles of Drosophila results in severely distorted, often furcated bristles, accompanied by disorganized actin bundles. We also fused the fli-I gene to green fluorescent protein (GFP) to be able to monitor its subcellular targeting using laser confocal microscopy. We assess the subcellular distribution of the protein relative to the structure and disposition of actin filaments, which are visualized using the F-actin specific fluorescent dye Alexa-phalloidin. Our results so far are consistent with a role of fli-I in the regulation of F-actin assembly, and "capping" of newly polymerized actin filaments. We are now investigating possible modifying effects of other regulatory genes, which would provide evidence for either antagonistic or synergistic interactions with fli-I. We believe that the Drosophila sensory bristle may be a convenient and sensitive in vivo system for the functional analysis of cytoskeletal regulatory proteins and the effects of pharmacological agents on cytoskeletal organization.
Basic Light and Fluorescence Microscopy Workshop

Wednesday, May 28, 2003
1:00 – 5:00 p.m.
University of Hawai‘i at Mānoa
Snyder Hall Rm 102 (lecture) and 115 (instruments)

Bob Nazar and Dennis Donley from Olympus America and Thomas Spieker with Soft Imaging Systems will be here for a half-day microscopy workshop at the Pacific Biomedical Research Center’s Biological Electron Microscope Facility.

Times below are approximate.

1:00 - 2:00  Comprehensive light microscopy, including brightfield, darkfield, phase, polarization, differential interference contrast and fluorescence. Filters, micrometers, reticles, troubleshooting and maintenance will be covered. Upright, inverted, and stereo microscopes will be available for hands-on demonstrations.

2:00 - 2:30  Presentation on total internal reflectance microscopy (TIRFM).

2:30 - 2:45  Principles of confocal microscopy.

2:45 - 3:30  Principles of digital cameras and image acquisition.

The remainder of the afternoon will be open for hands-on demonstrations of upright, inverted, stereo and TIRF microscopes, plus at least 4 digital cameras will be available for imaging of samples. A software specialist will be available to go over individual image analysis and processing needs, including deconvolution.

Refreshments will be served.

We encourage researchers, faculty, staff and students to attend to brush up on their microscopy skills!

If you are interested in attending, contact Tina Carvalho at the Biological Electron Microscope Facility by phone (956-6251) or email (tina@pbrc.hawaii.edu) so that we will have an accurate head count.