

THE MICROTOME

University of Vermont
College of Medicine



Microscopy Imaging Center Launches Newsletter

The MIC staff would like to welcome you to the premier issue of "THE MICROTOME". We have initiated this quarterly newsletter to keep you informed of happenings in the MIC. We will provide up-to-date information on the latest technical additions, and introduce new members of the facility. We welcome your input and suggestions for materials to be included in future issues!

Microscopy Imaging Center Receives Accreditation

We proudly announce that our laboratory was awarded accreditation, *with distinction*, by the College of American Pathologists (CAP). The CAP's program is highly respected and considered the "gold standard" of laboratory accreditation programs. This accreditation was based on the results of an on-site inspection in March and is designed to determine how well the laboratory serves the patient.



MIC technicians Michele von Turkovich and Janet Schwarz

tinely processes clinical biopsy material for electron microscopic study from patients at FAHC and outside facilities and is therefore subject to this evaluation. The inspectors examine the adequacy of the facility and its equipment, laboratory management and safety, qualifications of the staff, record keeping, and quality control.

The on-site inspection occurs every two years. Preparation for the inspection is an on-going process throughout the year but much last minute re-checking

of documentation and facility "polishing" occurs before the inspectors arrive.

The Microscopy Imaging Center rou-

Laboratory Tours

This summer, the Microscopy Imaging Center hosted several tours of our facility.

On May 21, eleven HELIX students were introduced to various microscopes and some of their applications. HELIX is a program at UVM dedicated to supporting undergraduate students in the sciences.

From June 25-28, the Microscopy Imaging Center participated in the Vermont Governor's Institute for Science and Technology. Thirty six students from the course brought in specimens collected from 3 different ecosystems in the area and viewed them in the scanning electron microscope with the assistance of Michele von Turkovich. This is a 4 day involvement for the Microscopy Imaging Center.

On August 14, fifteen people from the Vermont Genetics Network visited the MIC and were introduced to the various microscope technologies and some of their applications.

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- Jeol 1210 STEM
- Jeol T300 SEM
- BioRad MRC 1024 Confocal LSM
- Eppendorf Microinjector System
- DI Atomic Force Microscope
- Arcturus PixCell II LCM
- Zeiss Axioplan 2 Microscope
- CompuCyte Laser Scanning Cytometer
- Olympus BX50 Microscope Universal Imaging MetaMorph WS
- Dell NT Image Processing Workstation
- Morphologic services and consultation at the light and electron microscopy level.
- Morphometry (semi-quantitative morphology).
- Light and electron microscopic immunocytochemistry.
- Confocal scanning laser microscopy.
- Laser scanning cytometry.
- Atomic Force microscopy.
- Scanning electron microscopy.
- Laser capture microdissection. Preparation of paraffin sections and frozen sections.
- Training for use of the above equipment.
- Staining of sections with Picrosirius red.
- Testing of new antibodies, developing new staining techniques.
- Laser printing for publications and posters, computer-assisted digital imaging and analysis.



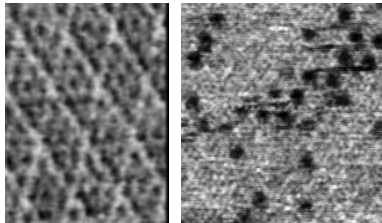
May The Force Be With You!

Atomic force microscopy is progressing in leaps and bounds at the College of Medicine's Microscopy Imaging Center. Doug Taatjes, Tony Quinn, Ted Bovill and collaborators Jacob Rand and Xiao-Xuan Wu of Mount Sinai College of Medicine have submitted a manuscript entitled: **Human Monoclonal Antiphospholipid Antibodies Disrupt Annexin A5 Binding to Phospholipid Bilayers: Evidence From Atomic Force Microscopy, Ellipsometry and Functional Assay.**

The study corroborates the hypothesis that antiphospholipid (aPL) antibodies can disrupt annexin A5 binding and crystalline lattice formation to phospholipid membranes and permit increased generation of thrombin.

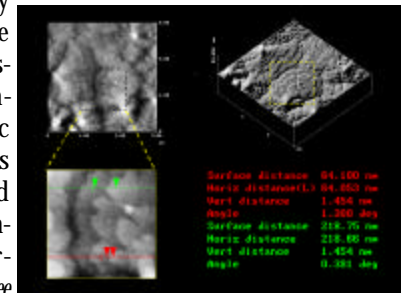
Lori Nylan of Molecular Biophysics and Physiology is working on a project under the direction of David Maughan & Jim Vigoreaux. As a former user of this pioneering technology, Lori returns to our facility to utilize the atomic force microscope to investigate mutant strains of *Drosophila* flightin null and mouse MyBP-C null lines to verify whether the thick filament diameter is significantly greater than that of wild type due to the loss of the thick-filament associated protein. They will also test the hypothesis that the hydrated filaments can be osmotically compressed by a high molecular weight dextran.

Presently, as with above imaging projects, all biological imaging is performed in fluid at ambient room temperature utilizing the AFMTM (atomic force microscopy tapping mode). We are working with IMF (Instrumentation & Manufacturing Facility) to design, develop and construct a perfusion chamber to provide precision temperature control, consistent volume levels, and stable component concentrations.



AFM images typically acquired are the high resolution image of intact annexin A5 shield (left) and of disrupted annexin A5 (right); dark areas are exposed lipid membrane.

Others utilizing our afm setup venture from across campus. Professor Randy Headrick and graduate students of the UVM Physics Department along with collaborator George Malliaras of Cornell University, Department of Materials Science will image surfaces of organic semiconductor thin films in air via tapping mode to investigate growth kinetics of such films. Organic semiconductor devices have experienced tremendous leaps in performance during the last 15 years. Light emitting diodes from organic semiconductors (OLEDs) are already produced in cell-phone displays and are about to enter the market in laptop displays. Studying the growth kinetics of organic semiconductor thin films using AFM as well as a variety of other techniques, including real-time xray scattering, will provide valuable physical measurements when combined with electronic transport measurements in transistors fabricated from organic semiconductor thin films. Surface measurements (see figure) will provide a more definitive understanding of organic growth and surface structures; and in turn, aid in improving design and higher precision fabrication of the films.



As indicated by the above and in the expanding literature, the field of atomic force microscopy is increasingly becoming another common modality in many fields of study to investigate both biological and material surfaces, providing both qualitative as well as quantitative information from cell surface tensile strength to material surface hardness. For more information on AFM see Tony Quinn or Doug Taatjes.

Goodbye Wendy!



At the end of September, Wendy Tra, our Dutch student returned home to Holland. Wendy has been a tremendous asset to our imaging center, completing an imaging study on compositional analysis of IRS-1 mice. She will be sorely missed! Thanks Wendy, for spending this year with us! We send you off with our best wishes for success.

Hello Nicole, Maria & Masha!

The Imaging Center would like to announce the arrival of three new characters. Maria Holton, UVM 2002 Graduate, is a Pathology graduate student. Nicole DeLance, UVM 2002 Graduate, is MIC's newest technician. Masha Stern is a technician working on the Imaging Core for Cell Signaling Program Project Group.

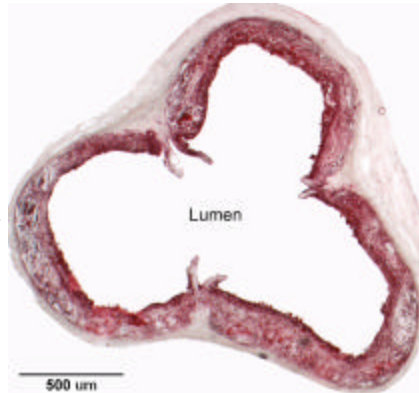


Microscopy & Microanalysis Conference in Quebec City

At the Microscopy Society of America international meeting held in Quebec City in August, **Marilyn Wadsworth** received the Biological Professional Technical Staff Award for 2002.

She presented a seminar entitled "Compositional Analysis of Atherosclerotic Lesions in a Mouse Model: A Validation of a new method that uses bright field, fluorescence and polarized light microscopy in conjunction with computer assisted image analysis to assess lesion composition."

Detailed methodology can be found in Wadsworth et al, *Delineation of the Evolution of Compositional Changes in Atheroma*, published in *Histochemistry and Cell Biology* in June 2002, Volume 118, pages 59-68.



Compositional Analysis of Atherosclerotic Lesion on a Mouse

MIC technician **Jan Schwarz** recently made a thirty minute presentation on "Project MICRO in Vermont"



at the Microscopy Society of America annual meeting in Quebec City, Quebec, Canada. The audience included both U.S. and international microscopists and educators and was intended as an exchange of ideas between programs currently in use. The presentation included a brief history of the three years that scientists at the University of Vermont have been active in Project MICRO (1230 students in 20 schools), contact strategies for "getting into" the schools, and ways to recruit

volunteers to staff the microscopic festivals. Logistical problems and solutions were highlighted, along with various venues and formats that have proven successful and the audience was invited to "sample" two learning stations that had been assembled. Jan concluded her presentation regarding some future directions of Project MICRO in Vermont, including pursuing alternative funding sources, and stimulating interest among the other colleges in the state to present programs in their areas.

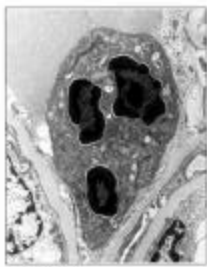
Please, see Jan Schwarz for more information about ProjectMICRO or to become a ProjectMICRO volunteer!



Middle Schools students participating in Project Micro

RECENT PUBLICATIONS:

- **Biotechniques:** *Laser-Based Microscopic Approaches: Application to Cell Signaling in Environmental Lung Disease.* Taatjes et al.(2001) 31:880-894
- **American Journal of Pathology:** *Increased Localization and Substrate Activation of Protein Kinase C delta in Lung Epithelial Cells Following Exposure to Asbestos.* Lounsbury et al. (2002) 160:1991
- **Cell Biology International:** *Structure and Dynamics of the Fusion Pore in Live Cells.* Cho et al. (2002) 26, 1, 35-42
- **Microscopy and Analysis:** *Microscopy-Based Imaging of Pathogenesis of Cardiovascular Disease.* Taatjes et al. (2001)
- **Thrombosis and Haemostasis:** *Time and Dose Dependent Augmentation of Inhibitory Effects of Abciximab by Aspirin.* Schneider et al. (2001) 85: 309-313
- **Histochemistry and Cell Biology:** *Delineation of the Evolution of Compositional Changes in Atheroma.* (2002) 118: 59-68
- **American Journal of Kidney Diseases:** *Biphasic Effects of Hemodialysis on Platelet Reactivity in Patients with End-Stage Renal Disease: a Potential Contributor to Cardiovascular Risk.* Aggarwal et al. (2002) 40, 2: 315-322
- **Experimental Gerontology:** *Imaging Techniques Used for the Detection of 8-Oxyquanine Adducts and DNA Repair Proteins in Cells and Tissues.* Persinger et al. (2001) 36(9): 1483-1494
- **Cancer Research:** *A Mutant Epidermal Growth Factor Receptor Targeted to Lung Epithelium Inhibits Asbestos-Induced Proliferation and Proto-Oncogene Expression.* Manning et al.(2002) 62: 4169-75



*Sometimes microscopy makes you scream!!!
by Janet Schwarz*

ATTENTION OLYMPUS/MAGNIFIRE USERS!

MetaMorph has now been relocated to a new computer in Room 203B. Olympus users requiring digital capture only need to sign up on one sheet. Please, see MIC staff for further details.

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<http://www.uvm.edu/microscopyimaging>



MIC Services Provided:

- Morphologic services and consultation at the light and electron microscopy level.
- Morphometry (semi-quantitative morphology).
- Light and electron microscopic immunocytochemistry.
- Confocal scanning laser microscopy.
- Laser scanning cytometry.
- Atomic Force microscopy.
- Scanning electron microscopy.
- Laser capture microdissection. Preparation of paraffin sections and frozen sections.
- Training for use of the above equipment.
- Staining of sections with Picosirius red.
- Testing of new antibodies, developing new staining techniques.
- Laser printing for publications and posters, computer-assisted digital imaging and analysis.

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