CourseTopics:

Tissue processing for EM, immunolabeling and the fundamentals of tissue processing for paraffin.

Hands on Techniques:

Tissue processing for EM and immunolabeling.

Location:

Life Sciences Microscopy Facility at Purdue Univ. Whistler Hall of Agriculture Research - Room S-052; 170 South University Street; West Lafayette, IN

The Life Science Microscopy Facility at Purdue University is a core facility located in the Whistler Research Building. This 4000 sq. ft. laboratory complex supports research on microscopic and submicroscopic structure of biological and physical systems and is available to the campus community. Technology in the LSMF provides capabilities for transmission electron microscopy with EDX, STEM, HAADF and tomography, field emission scanning electron microscopy with cryo and EDX and a dual-beam instrument with cryo and EDX. Equipment is available for cryo sample preparation (high pressure freezing and freeze substitution, ultramicrotomy, critical point drying, vacuum evaporation and sputter coating. Staff in the LSMF provide expertise in a wide range of specialized preparation techniques including immunocytochemistry and freeze substitution to assist researchers. The LSMF is undergoing a merger with the Biological EM facility which will add 3 cryo TEMs including a Titan Krios and a CM300FEG in a BSL3 facility.

Directions and Parking:

From Chicago, Fort Wayne, Indianapolis, South Bend:
Exit I-65 at Exit 172 and proceed west (toward Lafayette) on South Street. Proceed four miles to West Lafayette. When you cross the Wabash River you are in West Lafayette and are now on State Street. Past the fourth traffic light and turn left on Sheetz Street. See below for on-campus directions to facility.

From Champaign, Crawfordsville, St. Louis, Terre Haute:
Exit I-70 (exit 41) or I-74 (exit 34) on U.S. 231 North for 54 miles. When you cross the Wabash River, you are in West Lafayette. Turn right on South River Rd., proceed 1.3 miles, then turn left on IN-26/State Street. Make slight left onto West State St., then turn left on Sheetz St. See below for on-campus directions to facility.

On-Campus Directions to Facility:
The Marsteller Parking Garage (PGM on campus map) will be ½ block on your right after turning onto Sheetz St. From this parking garage, Whistler Hall is two blocks west on South University Street. The address is: Whistler Hall, 170 S. University St. The Life Sciences Microscopy Facility is located in Rm. S-052 – take elevators or stairs to basement at either end of the building.

Parking:
Visitors may purchase a daily permit from the Purdue Parking Facilities website: https://purdue.t2hosted.com/cmn/index.aspx. A daily permit allows visitors to park in any of the parking garages normally reserved for "A" permits. Parking is also available at the street parking meters ($0.25/15 min.), but not recommended.

Fig. A-B shows cytoplasmic iridovirus from the skin of a sturgeon. The iridovirus is a large enveloped dsDNA virus which infects both insect and vertebrate hosts. Fig. C-E demonstrates anintranuclear baculovirus from the hepatopancreas of a crayfish from Northern California. Fig. C is a low magnification image of the enveloped dsDNA virus showing the intranuclear arrangement
of virus particles Fig. D is a higher magnification showing both a cross-sectional and longitudinal view of the virus. Fig. E is a high magnification cross-section of a number of virus particles demonstrating the unique intranuclear membrane-bound virions. Infected tissues were processed directly from 10% NBF by the microwave methods of Nordhausen and Barr (2001) and Nordhausen et al. (2002).

Nordhausen RW, Barr BC, Hedrick RP (2002) Microwave-assisted rapid tissue processing for disease diagnosis in a veterinary diagnostic laboratory. Microsc Microanal 8(Suppl 2):150-151

Nordhausen RW, Barr BC (2001) Specimen preparation for thin-section electron microscopy utilizing microwave-assisted rapid processing in a veterinary diagnostic laboratory. In Giberson RT, Demaree RSJr, eds. Microwave Techniques and Protocols, Totowa, NJ, Humana Press, 49-66

Retinas were fixed in 4% paraformaldehyde in 0.1M phosphate buffer (pH 7.4) overnight at 4°C. Following labeling the tissue was rinsed 6 x 20 minutes in buffer prior to beginning antibody labeling. The bench labeling protocol requires 7 days. These labeling results were completed in an afternoon using microwave-enhanced labeling (PELCO BioWave® with SteadyTemp™) during a workshop held at the Univ. of Minnesota Imaging Center (Mark Sanders, Director - May 17-19, 2006). The retinas were double-labeled for:

- Collagen Type IV (basal lamina surrounding blood vessels) with rabbit anti-type IV collagen and the secondary conjugated to FITC (green label)
- Glutamine Synthetase (enzyme found in retinal Müller glial cells) with mouse anti- glutamine synthetase and the secondary conjugated to Cy3 (red label)

Primary antibody labeling was done at 170Wfor 12 minutes (4 on - 4 off - 4 on) under vacuum (15" Hg). Secondary antibody labeling was done at 170W for 6 minutes (2 on - 2 off - 2 on) under vacuum (15" Hg). Images were collected on a Nikon C1si Confocal Microscope.

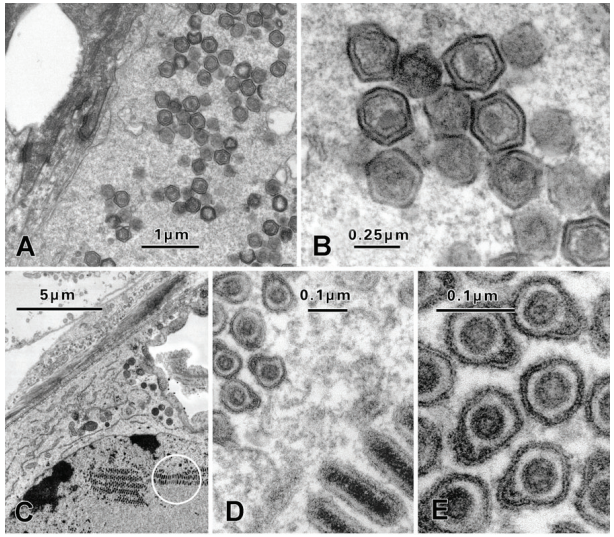
Microwave Workshop

Presented by the

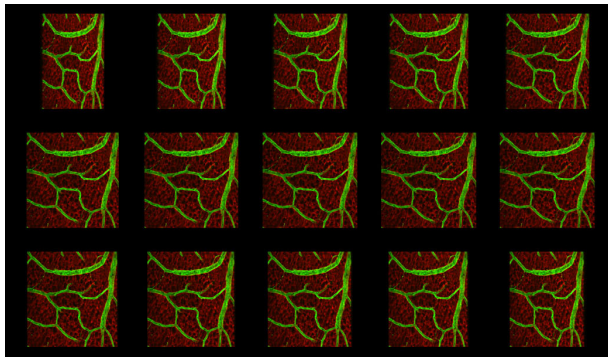
Life Sciences Microscopy Facility, Purdue University and Ted Pella, Inc.

April 15 – 17, 2014

Electron Microscopy



Immunolabeling



Course Outline:

Tuesday, April 15th

8:30am – 12:00pm (Lecture)

- Introduction
- Understanding Microwave Technology
- Overview of Applications
- Tissue Processing for EM - Theory and Application Fundamentals

12:00 – 1:00pm (Lunch)

1:00 – 5:00pm (Hands-On Session)

- Tissue Processing for EM
- Review, Q&A, Wrap-Up

Evening Free

Wednesday, April 16th

8:30am – 12:00pm (Lecture)

- Finish Tissue Processing for EM
- Processing Difficult Tissues

12:00 – 1:00pm (Lunch)

1:00 – 4:30pm (Hands-On Session)

- Finish Tissue Processing for EM
- Immunolabeling Technique
- Review, Q&A, Wrap-Up

6:30 – 9:00pm (Group Dinner)

Thursday, April 17th

Start 9:00am – 12:00pm (Lecture)

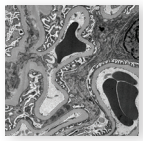
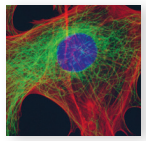
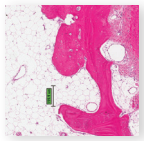
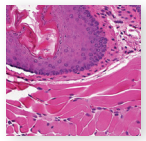
- Finish Hands-On Sessions
- Evaluate Results

12:00 – 2:00pm (Lunch/Discussion)

- Q&A, Wrap-Up, Course Evaluation

Course Includes:

April 15th dinner will be responsibility of participant, April 16th dinner provided – please indicate special requirements.

Confirmations:

Confirmations will be sent out immediately on receipt of the registration form.

Contact Information:

Kathy Stangenberg
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Email: sales@tedpella.com
Phone: 800-237-3526 ext. 201

Jerry Jasso
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Phone: 330-319-4236

Chris Gilpin, Ph.D.
at Life Sciences Microscopy Facility,
Purdue University
Email: gilpin@purdue.edu
Phone: 765-494-7750

Number of Participants:

The workshop is limited to 12 participants. Early registration is recommended. Six participants required to hold the workshop. Registration fees will be refunded if the workshop fails to achieve the requred number of participants.

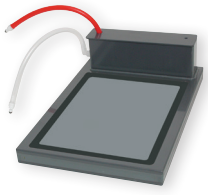
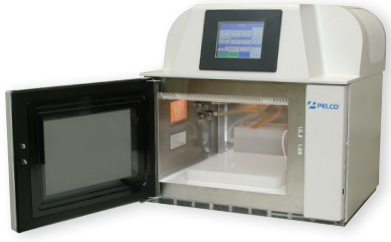
Cancellations:

No cancellations will be accepted within 2 weeks of the workshop date.

Lodging:

Campus Inn
200 Brown St, West Lafayette, IN 47906
Phone: (765) 743-9661
Rate: $50.00/night plus tax;
Mention "Microwave Workshop rate from Debbie".
Cancellation policy 24 hours prior to stay.

Other hotels include Hilton Garden Inn, University Plaza Hotel, Holiday Inn Lafayette at City Centre.

Microwave Workshop Purdue University Registration Form:

Name _____

Address _____

_____

_____

_____

Phone _____

FAX _____

Email _____

Payment:

Registration Fee is $500.00

- ☐ VISA
- ☐ Master Card
- ☐ American Express
- ☐ Payment Enclosed
- ☐ Purchase Order

Card No. _____

Expiration Date _____

Make all registration fees payable to:
Ted Pella, Inc.

Return form to:
Kathy Stangenberg
Ted Pella, Inc.
P.O. Box 492477
Redding, CA 96049-2477

FAX: 530-243-3761

Phone: 800-237-3526 ext. 201

Register no later than April 1, 2014.