BIOB 596: ST Principles of Light Microscopy

Class: 2:30 – 4:00 pm; Monday & Thursday, HS108
Prof: Dr. Scott Wetzel (SC216, ph: 243-2168, scott.wetzel@mso.umt.edu)
Textbook: Handbook of Biological Confocal Microscopy, 3rd Ed., James Pawley, editor

Date	Topic	Reading
	January	
23	Introduction, Basic Optics: Refraction and Diffraction	
26	Basic Optics: Diffraction and Resolution	20, p36-39
30	Basic Optics: Contrast Formation	4, 8
	February	
2	Fluorescence and Contrast Formation	8 (p172),
6	Optical Filters and Lenses	7, p43-50, 54-6
9	Detectors	Appendix 3, ch12, p233-5
13	Fluorescent Proteins	
16	Fluorescent Probes : Dyes, Photobleaching & Ions	16, 17, 39, 42
20	**HOLIDAY**	
23	Imaging Modality Comparison	1, 23
27	Wide-field Microscopy	23
	March	
2	Deconvolution	
6	Basic Confocal Microscopy	36
	March	
9	Spinning Disk and Multi-photon Imaging	10
13	Multi-photon Imaging	
15	**MID TERM EXAM**	
20 & 22	**SPRING BREAK – No Classes**	
27	TIRF	
29	Super Resolution: Structured Illumination, STED, PALM, STORM	31
	April	
3	Super Resolution: Structured Illumination, STED, PALM, STORM	31
5	Advanced Techniques: FRET, FRAP, FLIM	45, 27
7	Advanced Imaging Techniques: Light Sheet	37
10	Live Cell Imaging	19
12	Experimental Design and Specimen Prep	18
17 &19	***No Scheduled Class – Hands-on experience Times to be arranged individually***	35, 36
24	Digital Images and Image Processing	14
26	Image Processing: Filters	48
	May	
1	Image Processing: Colocalization	p 667-70
3	Presentation and Publication	48

BIOB 596: ST Principles of Light Microscopy

Course Overview: Imaging is a powerful technique and over the past decade it has become an important component of cutting-edge research in many areas of biological research. This course is designed to provide students with an understanding of current light microscopy techniques. This will help students design and carry out their own imaging experiments and critically assess imaging data presented in the literature. This will be achieved through a combination of lecture-based presentations and several hands-on technical sessions

GRADING will be based on the following:
100 points – Midterm Exam
100 points – Final Exam.
50 points – Data analysis problems
100 points – Research Article Review
Total Points Possible = 350 points

Final grades will be based upon a straight 10% grading scale based upon the total number of points (90% for A, 80% for B, 70% for C, 60% for D, below 60% = F). Late policy is outlined below.

Late Work:

Late work is strongly discouraged. **For assignments with a specified due date, a late penalty of 10% per day of tardiness will be subtracted from the grade**.

Exams: Excuses for rescheduling or missing and exam must be approved BEFORE the exam. If not pre-approved, no makeup exam will be given and a grade of F (a score of 50%) will be recorded for the exam. Anyone missing the final exam will receive a grade of F for the entire course.

Important Note: Cheating on an exam will result in a grade of ZERO for that exam.

Supplemental Reading:

While the textbook provides a very good resource, from time to time review articles will be assigned that cover special topics. These articles will be made available on the Moodle web page for this class.

Web-based Resources:

The following are excellent web resources for imaging concepts and techniques:
http://micro.magnet.fsu.edu/primer/
http://www.olympusmicro.com/primer/virtual/virtual.html

Students should download ImageJ and plugins from http://rsb.info.nih.gov/ij/ or Fiji https://fiji.sc.
These are cross-platform java-based image analysis programs. They're powerful programs and they're FREE!

There is a very active and excellent confocal mail list at
http://listserv.buffalo.edu/archives/confocal.html

Hands-On Experience:

It is essential to a comprehensive understanding of light microscopy to get some hands-on experience with imaging techniques. Towards that end, there are no classes scheduled for the week of April 17. Instead, students will prepare samples and image them on the confocal system on campus. The day and time will be individually arranged with the instructor. Samples can be provided, but students are highly encouraged to prepare samples from their own research projects to image.

Data Analysis Problems:

DUE DATE: Final Exam

Students will be given several image files to analyze the first week of April. We will be discussing image processing and presentation during April to introduce students to image processing techniques. Students will perform the requested image analysis using ImageJ, Fiji, or a proprietary imaging software package. The requested results will be due the day of the final exam.

Research Article Critique:

DUE DATE- Fri., May 5; 5:00 pm

Directions: Choose a 2011-20015 **primary research** article that contains at least 2 imaging figures in a field that interests you, read it thoroughly, and write a review. This review will not be a typical synopsis of the article. Rather, the purpose of this exercise is to critique the imaging techniques employed and the results obtained.

Format:

- 4 Pages MAXIMUM double-spaced and typed. (less is better)

- This paper should include a brief (1 – 3 paragraphs) introduction to the content of the paper. Give an overview of the research question and their findings. Write it for someone outside of the particular field to provide context and so that they can understand the overall content of the paper.

- This should be followed by a thorough discussion of the imaging data in the paper. This should include the rationale for their imaging experiments, a description of their imaging techniques and the results. This should be critically examined –were the techniques appropriate, the results interpreted correctly, etc. What should they have included/changed? This section of the paper should be no more than 2.5 pages.

BIOB 596: ST Principles of Light Microscopy

Learning Outcomes:

Upon completion of this class, the student will have:

1. Gained experience in reading current primary biomedical/ biochemical scientific literature pertaining to the special topic described for the semester.

2. Gained a deeper understanding of the details of the state of the current research in light microscopy including wide-field and confocal microscopy and more specialized cutting-edge technologies such as superresolution, light sheet, and FLIM,

3. Gained experience in preparing and presenting scientific data to an audience that is moderately conversant in the area of study.

Accessibility, disabilities, and special accommodations:

The University of Montana assures equal access to instruction through collaboration between students with disabilities, instructors, and Disability Services for Students (DSS). If you think you may have a disability adversely affecting your academic performance, and you have not already registered with DSS, please contact DSS in Lommasson 154. I will work with you and DSS to provide an appropriate accommodation.

Please note: You are bound by the University of Montana student conduct code. All work will be performed solely by the student. Plagiarism and cheating of any kind will result in referral for disciplinary action and you will receive a zero on the assignment. This will significantly impact your final course grade.