

ANNB328- Techniques in optical microscopy

Instructors:

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This team-taught course will serve as an introduction to many of the optical microscopic techniques available at the UVM College of Medicine. The course format consists of lectures, literature, discussions and demos. Topics include general light microscopy and contrast techniques, as well as epifluorescence, confocal, multi-photon, and post-processing (deconvolution) techniques. Applications (FRET, ion imaging, second-harmonic generation imaging, FRAP, TIRF, FLIM etc.) will also be discussed as time permits.

It will meet twice a week: Monday and Wednesday, from 1:30-3:00 pm in HSRF 400.

Date	Topic 1	instructor	Date	Topic 2	Instructor
1/14	Introduction + light properties Diffraction/ refraction, polarization	Clason	1/16	Basic Scope/ Kohler, resolution, aberrations - part 1	Taatjes
1/21	MLK Day – no class		1/23	Basic Scope/ Kohler, resolution, aberrations – part 2	Taatjes
1/28	Polarized microscopy, Phase, Hoffman, DIC Catch-up/ demo	Taatjes	1/30	Dye chemistry- absorbance, emission, excitation, bleaching, uncaging	Clason
2/4	Lamps, lasers, filters, fibers monochromators, epi-scope design	Fiekers	2/6	Fluorescence detectors- CCD, PMT, noise	Taatjes
2/11	Sample preparation techniques: immunohistochemistry, live cells, fixation methods, transfection	Fiekers	2/13	Epifluorescence Papers + demo	Fiekers + Taatjes
2/18	President’s Day – no class		2/20	Confocal + spinning disk	Taatjes
2/25	Confocal papers + demo	Taatjes	2/27	TIRF	Clason
3/3	TIRF papers + demo	Clason	3/5	Multiphoton confocal- Including SHG	Clason
3/10	Spring recess – no class		3/12	Spring recess – no class	
3/17	Multiphoton papers + demo	Clason	3/19	Post-processing deconvolution	Clason
3/24	Deconvolution papers + demo	Fiekers + Clason	3/26	Calcium/ ion imaging Ratiometric, non-ratiometric	Fiekers
3/31	Ion imaging papers + demo	Clason	4/2	FRET	Fiekers
4/7	FRET papers + demo	Fiekers	4/9	Spectral Detection	Taatjes
4/14	Spectral papers + demo	Taatjes	4/16	FLIM	Clason
4/21	FLIM papers	Clason	4/23	FRAP/ FLIP	Clason
4/28	FRAP papers + demo	Clason	4/31	Final Exam	

Grading:

50% of the grade will be based on class preparedness and participation

50% will be based on exams.

Discussion sections:

Students will present papers from our collection, or of their choosing on the current topic.

Please limit your paper choices to topics that have been discussed already and have your selection ready one week before your scheduled date. That will give us all a chance to photocopy and read the paper. This is a techniques-based course, so the presentations should focus mainly on the methods used and should reflect on the following questions:

1. What technique was used?
2. Was the technique well-suited to the question they were asking?
3. Was the technique well-suited to their sample type?
4. Were the proper controls done?