**Biology 542 –Syllabus –Fall 2019**

**Light Microscopy for Biology and the Biomedical Sciences**

**Professors:** P.S. Maddox and Kerry Bloom

**Course TA**: Tanner Fadero ([fadero@live.unc.edu](mailto:fadero@live.unc.edu))

**Class meeting**:  Tues, Thurs: 4:45 – 6:00, TBA

Resources

Microscopy Book: Fundamentals of Light Microscopy and Electronic Imaging, Douglas Murphy, Wiley-Liss, 2001 (available on Sakai web site)

Handouts in Class: TBA

Grades

Grading will be based on ~10 weekly homework assignments (10 pts each) a take home exam (100 pts.) as well as individual or group independent projects (200 pts).

Schedule

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| **Date** | **Weekday** | **Topic** |  | **Instructor** |  |
| Aug 11 | Tuesday | Introduction |  | Maddox, Bloom, Fadero |  |
| Aug 13 | Thursday | Introduction: Discussion of life inside a cell |  | Bloom |  |
| Aug 18 | Tuesday | Reflection, Refraction, Interference |  | Bloom |  |
| Aug 20 | Thursday | The Origins and Evolution of Light Microscopy for Biology |  | Bloom |  |
|  | Reading assignment for Sept 10: *Fundamentals of light microscopy and electronic imaging* pages 1-13 (Basic illuminating light paths, conjugate image planes) and pages 15-50 (Geometrical optics and basic imaging light paths) | | | | |
| Aug 25 | Tuesday | Geometrical optics and basic imaging light paths |  | Bloom |  |
| Aug 27 | Thursday | Microscope assembly, alignment, identification of major components, identification of conjugate image planes |  | Bloom |  |
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| Sept 1 | Tuesday | Rails Lab– Lenses and Focal Length |  | Tanner |  |
|  | Reading assignment for Sept 12: <https://www.microscopyu.com/tutorials/kohler> <https://www.microscopyu.com/microscopy--‐basics/components> | | | | |
| Sept 3 | Thursday | Rails Lab – Multi-­‐lens systems |  | Tanner |  |
|  | Reading assignment for Sept 17: *Fundamentals of light microscopy and electronic imaging* pages 117-133 (Polarized light interaction with matter) | | | | |
| Sept 8 | Tuesday | ***Label free microscopy and proper Kohler alignment***. Emily and Tanner  Including: Brightfield, DIC, and Phase Contrast |  | Tanner |  |
| Sept 10 | Thursday | Polarization and DIC |  | Bloom |  |
| Sept 15 | Tuesday | Lab on Polarization |  | Tanner |  |
| Sept 17 | Thursday | Introduction to fluorescent probes, their fundamental properties and how they can be used as bio-sensors |  | Bloom |  |
|  | Take home exam 1 (due XX) | | | | |
|  | Reading assignment for Oct 1: Resolution and point spread function (A beginner’s guide to rigor and reproducibility in fluorescence imaging experiments by Lee et al) https://www.molbiolcell.org/doi/10.1091/mbc.E17-05-0276 | | | | |
| Sept 22 | Tuesday | Lab on Fluorescence |  | Tanner |  |
| Sept 24 | Thursday | Applications of Multi-wavelength GFP, YFP, CFP and DIC imaging in budding yeast GFP live cell biology |  | Bloom |  |
| Sept 29 | Tuesday | Lab on Fluorescence II |  | Tanner |  |
| Oct 1 | Thursday | Widefield vs Confocal comparison. Resolution and Point spread function A beginner’s guide to rigor and reproducibility in fluorescence imaging experiments |  | Maddox |  |
| Oct 6 | Tuesday | Lab exercise |  | Tanner |  |
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| Oct 8 | Thursday | Practical Applications, counting molecules, nanometer localization accuracy in wide-field |  | Maddox |  |
| Oct 13 | Tuesday | Intro. Super-Resolution Microscopy: SHREC, SIM, PALM, STORM |  | Maddox |  |
|  | Reading assignment for Oct 29: Intro. Video and Digital Cameras, digital images, digital image acquisition, microscope control: A Multi-Mode Digital Imaging Microscope (MetaMorph and Image J; 236-238; 260-267; Image J Instructions) | | | | |
| Oct 15 | Thursday | Introduction to video and digital cameras, digital images, and digital image acquisition |  | Maddox |  |
|  |  | Reading assignment for Nov 7: Point Scanning Confocal and Multi-photon Microscopy: Basic concepts and practical aspects (205-231) |  |  |  |
| Oct 20 | Tuesday | Point Scanning Confocal and Multi-photon Microscopy: Basic concepts and practical aspects (205-231) |  | Maddox |  |
| Oct 22 | Thursday | Getting quantitative information from photon counting to digital images and digital image processing (259-281) |  | Maddox |  |
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| Oct 27 | Tuesday | 3-D Image De-Convolution |  | Maddox |  |
|  | Reading assignment for Nov 12: Getting quantitative information from photon counting to digital images and digital image processing (259-281) | | | | |
| Oct 29 | Thursday | Advanced Fluorescence Methods: FRAP, FLIP, FRET, TIRF |  | Maddox |  |
| Nov 3 | Tuesday | Examples of Single Molecule Imaging of protein function with TIRF Examples of FRET BioSensors: phosphorylation, tension |  | Maddox |  |
| Nov 5 | Thursday | Cell based screening for identifying functional proteins |  | Maddox |  |
| Nov 10 | Tuesday | Real world examples: single molecule imaging for protein function using TIRF; FRET biosensors for measuring phosphorylation, tension, etc |  | Maddox |  |
| Nov 12 | Thursday | Cell based screening for identifying functional proteins |  | Maddox |  |
| Nov 17 | Tuesday | Independent projects |  |  |  |
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