Course Documents

Syllabus
Lecture and Presentation Schedule (this just shows topics, specific readings will be posted below)
Major Writing Assignment: Mini NIH-Style Grant Proposal
   For guidance on what goes in each section and the criteria by which your proposal will be judged, consult these excerpts from the NIH's instructions.
   (The entire instructions can be found here.)
   Here are some examples of Specific Aims pages from recent proposals: Hutson, Gochberg.
   I would also suggest consulting this excellent guide to scientific writing in general: "The Science of Scientific Writing" by George D. Gopen and Judith A. Swan. If you find that interesting, but need more, Gopen has an excellent book available entitled The Sense of Structure: Writing from the Reader's Perspective.
Course blog for answering Warm-Up questions

This Week's Reading

Monday, December 12 at 9:00 AM - Proposals DUE - Please submit ELECTRONICALLY to shane.hutson@vanderbilt.edu

Documents to provide you with some guidance in preparing your proposals
1. Instructions: Major Writing Assignment: Mini NIH-Style Grant Proposal
2. For guidance on what goes in each section, consult these excerpts from the NIH's instructions. (The entire instructions can be found here.)
3. For details on the criteria by which your proposal will be judged, see the NIH's Research Program Grant Critique Template (editable Word .doc), critique template instructions and an NIH-provided example critique (which also has links to descriptions of all the review criteria). You will notice that I have crossed out the Investigator and Environment criteria in the template and example. You will not be evaluated on these criteria.
4. Details and guidance on the NIH scoring system.
5. Everyone should read this very short site from the Vanderbilt library on Avoiding Plagiarism. For more information, you can follow links from the Vanderbilt Writing Studio's Tips on Avoiding Plagiarism.

Sample Proposals:
1. My last NIH proposal (a bit longer than what I'm asking of you, but it was funded $$)
2. Best-scored student proposal from Fall 2009: Lewis Kraft
3. Best-scored student proposals from Spring 2008: Carly Anderson and Ken Lewis
4. Best-scored student proposal from Spring 2006: Zhemin Wu

http://www.vanderbilt.edu/lsp/B325/
Thursday, December 15 at 3:00 PM - Mock Study Section

Everyone should read the specific aims section of ALL proposals. In addition, you will each have TWO proposals for which you will write a formal review using the NIH's Research Program Grant Critique Template (editable Word .doc). To help you, NIH provides an example critique (which also has links to descriptions of all the criteria you are to consider) and critique template instructions. You will notice that I have crossed out the Investigator and Environment criteria in the template and example. Please ignore these criteria in your critiques.

In addition to the written critique, you will need to decide on scores (1-9, 1 being best) for each remaining criterion and for the overall proposal. The following link provides details and guidance on the NIH scoring system.

In our mock study section, the formal reviews of each proposal will be presented by the two assigned students and then the proposal will be discussed by the entire panel. We will all vote on the quality of the proposal (via a 1-9 score) and average these to come up with an overall priority score. At the end of the session, we will look back on the distribution of priority scores amongst all the proposals (to see who would get $$$).

I will send your review assignments via email. These reviews are single-blind - i.e. you know whose proposal you are reviewing, but you will not know who is reviewing your proposal.

Full Proposals
1. Bryson Brewer
2. Juan-Carlos Cordova
3. Monica Lacy
4. Nichole Lareau
5. Ty McCleery
6. Jessica Moore
7. Jie Zhao

Specific Aims of All Proposals

Previous Readings/Lecture Notes

Tuesday, August 30 - Introduction to Fluorophores and Fluorescence - WARM-UP QUESTION ON COURSE BLOG
Two sections from the Molecular Expressions website maintained by the Optical Microscopy Division of the National High Magnetic Field Laboratory (at Florida State). In addition to the sections you'll read for my lectures, this site is a great resource if you need background information for your own presentation on fluorescence techniques.
1. Basic Concepts in Fluorescence If fluorescence is completely new to you, you may want to start with Introduction to Fluorescence and Overview of Excitation and Emission Fundamentals.
2. Fluorophores for Confocal Microscopy

Thursday, September 1 - Confocal and Multiphoton Fluorescence Microscopy - WARM-UP QUESTION ON COURSE BLOG
1. Introduction to Confocal Microscopy (mostly laser scanning confocal) with an interactive java tutorial.
2. Introduction to Spinning Disk Confocal Microscopy with a flash tutorial on the Yokogawa design.

http://www.vanderbilt.edu/lsp/B325/
3. **Multiphoton Fluorescence Microscopy**

**Tuesday, September 6** - Fluorescence (or Förster) Resonance Energy Transfer (aka FRET) - **WARM-UP**

1. FRET - Introductory Concepts
2. Spectral Imaging and Linear Unmixing

**Thursday, September 8** - Super-Resolution Fluorescence Microscopy - **WARM-UP**

1. Introduction to Super-resolution Microscopy with links to several interactive tutorials that cover almost all the super-resolution techniques.

**Tuesday, September 13** - Student Presentations - **WARM-UP**


**Thursday, September 15** - Student Presentations - **WARM-UP**


**Tuesday, September 20** - Student Presentations - **WARM-UP**


**Thursday, September 22** - Student Presentations - **WARM-UP**


**Tuesday, September 27** - Introduction to Optical Tweezers - **WARM-UP** - PPT Slides

1. M.C. Williams, *Optical tweezers: Measuring piconewton forces*

**Thursday, September 29** - Measurement of Molecular & Cellular Forces - **WARM-UP** - PPT Slides


**Tuesday, October 4** - Student Presentations - **WARM-UP**


**Thursday, October 6** - Fall Break - No Class

**Tuesday, October 11** - Student Presentations - **WARM-UP**
Tuesday, October 11 - Student Presentations - WARM-UP

Thursday, October 13 - Vibrational Molecular Spectroscopy - WARM-UP - PPT SLIDES
1. Infrared Spectroscopy Primer by William Reusch of Michigan State University (one of several nice spectroscopy links at SpectroscopyNow)
2. Introduction to FTIR Spectroscopy from Oriel Instruments.
3. The Fourier Transform Toolkit - you don't have to read this beforehand, but it is a good reference for building intuition about Fourier Transforms (which we will use repeatedly in the spectroscopy and MRI sections). If you aren't really familiar with FTs, it might help you with the reading questions.

Thursday, October 18 - Vibrational Molecular Spectroscopy - PPT SLIDES
No new reading.

Tuesday, October 25 - Student Presentations - WARM-UP

Thursday, October 27 - Student Presentations - WARM-UP
2. <Jie> Sumii, M; Furutani, Y; Waschuk, SA; et al. Strongly hydrogen-bonded water molecule present near the retinal chromophore of Leptosphaeria rhodopsin, the bacteriorhodopsin-like proton pump from a eukaryote Biochemistry 44 (46): 15159-15166 (2005).

Tuesday, November 1 - Magnetic Resonance Imaging - WARM-UP
Ch 3, Spin Physics
Ch 6, Imaging Principles
Ch 7, Fourier Transform Imaging Principles

Thursday, November 3 - Student Presentations - WARM-UP

Tuesday, November 8 - Mass Spectrometry - WARM-UP

Thursday, November 10 - Mass Spectrometry (continued)
Thursday, November 10 - Mass Spectrometry (continued)
• "The Science of Scientific Writing" by George D. Gopen and Judith A. Swan
• ABSTRACTS for proposals are due.

Tuesday, November 15 - Student Presentations - WARM-UP
3. Don't forget that class will be extended to make-up for a previous mixed lecture. We'll spend this time discussing your abstracts in relation to "The Science of Scientific Writing" by George D. Gopen and Judith A. Swan.

Thursday, November 17 - Student Presentations - WARM-UP

Thursday, December 1 - Electrochemical Measurements - WARM-UP

Tuesday, December 6 - Student Presentations - WARM-UP

Thursday, December 8 - Student Presentations - WARM-UP

FTIR SPECTROSCOPY

Student Presentation Options

Liu, M; Krasteva, M; Barth, A Interactions of phosphate groups of ATP and aspartyl phosphate with the sarcoplasmic

http://www.vanderbilt.edu/lsp/B325/

Sumii, M; Furutani, Y; Waschuk, SA; et al. Strongly hydrogen-bonded water molecule present near the retinal chromophore of Leptosphaeria rhodopsin, the bacteriorhodopsin-like proton pump from a eukaryote. *Biochemistry* 44 (46): 15159-15166 (2005).


**MOLECULAR & CELLULAR FORCE MEASUREMENTS**

**Optical Tweezers**


Magnetic Tweezers and Other Techniques

NEW REVIEW ARTICLE - I wouldn't suggest presenting this specific article, but it has lots of great links to papers that could make very nice presentations. Brenner, M.D.; Zhou, R.; and Ha, T. *Forcing a Connection: Impacts of Single-Molecule Force Spectroscopy on In Vivo Tension Sensing* Biopolymers 95(5): 332-344 (2010).

Shroff, H; Reinhard, BM; Siu, M; Agarwal, H; Spakowitz, A; Liphardt, J; *Biocompatible force sensor with optical readout and dimensions of 6 nm* Nano Letters 5 (7): 1509-1514 (2005).

Charvin, G; Strick, TR; Bensimon, D; Croquette, V; *Tracking topoisomerase activity at the single-molecule level* Annual Review of Biophysics and Biomolecular Structure 34: 201-219 (2005).

Danilowicz, C; Greenfield, D; Prentiss, M; *Dissociation of ligand-receptor complexes using magnetic tweezers* Analytical Chemistry 77 (10): 3023-3028 (2005).


Tan, JL; Tien, J; Pirone, DM; Gray, DS; Bhadriraju, K; Chen, CS; *Cells lying on a bed of microneedles: An approach to isolate mechanical force* Proceedings of the National Academy of Sciences, USA 100 (4): 1484-1489 (2003).

Desprat, N; Richert, A; Simeon, J; Asnacios, A; *Creep function of a single living cell* Biophysical Journal 88: 2224-2233 (2005).

Wottawah, F; Schlinkinger, S; Lincoln, B; Ananthakrishnan, R; Romeyke, M; Guck, J; Kas, J; *Optical rheology of biological cells* Physical Review Letters 94: 098103 (2005).


**Laser Microdissection**


### FLUORESCENCE

**Super-resolution Microscopy**


*Pavani, SRP; Thompson, MA; Biteen, JS, et al. (2009) Three-dimensional, single-molecule fluorescence imaging beyond the diffraction limit by using a double-helix point spread function Proc Natl Acad Sci USA 106(9): 2995-2999.*


[http://www.vanderbilt.edu/lsp/B325/]


NEW Wang, YX; Botvinick, EL; Zhao, YH, et al. (2005) Visualizing the mechanical activation of Src Nature 434(7036): 1040-1045.


**FLIM (Fluorescence Lifetime Imaging Microscopy)**


**Single Molecule and Fluctuation Techniques**


**Genetically-encoded Fluorescent Molecules**


**Two-photon and Multi-photon Microscopy**


**Specialized Techniques**


NMR SPECTROSCOPY & MRI

Helpful Online Tutorials (Supplemental material)

"Understanding NMR Spectroscopy", James Keeler, University of Cambridge
http://www-keeler.ch.cam.ac.uk/lectures/Irvine/

"Nuclear Magnetic Resonance (NMR)" in Biophysics Textbook Online, David Gorenstein, Volume Editor
http://www.biophysics.org/btol/NMR.html

"The Basics of MRI", Joseph P. Hornack
http://www.cis.rit.edu/htbooks/mri/inside.htm

"Magnetic Resonance Imaging - A Window into the Human Body", Jim Feeney
http://www.nimr.mrc.ac.uk/millhillessays/1996/mri.htm

Student Presentation Options: NMR/EPR/ESR Spectroscopy


N. Tjandra, A. Bax (1997) "Direct Measurement of Distances and Angles in Biomolecules by NMR in a Dilute Liquid Crystalline Medium" Science 278: 1111-1114.


D. Hilger, Y. Polyhach, E. Padan, H. Jung & G. Jeschkey (2007) "High-Resolution Structure of a Na+/H+ Antiporter Dimer Obtained by Pulsed Electron Paramagnetic Resonance Distance Measurements" Biophysical Journal 93: 3675–3683. with New and Notable blurb by A.H. Beth "How to Assemble the Parts: Structures of Protein Complexes from their Components"


**Student Presentation Options: MRI and Functional MRI**


http://www.vanderbilt.edu/lsp/B325/


MICROFLUIDICS & CELLULAR INSTRUMENTATION

Student Presentation Option


This review article should not be selected for a presentation, but it does have lots of references that would make good presentation papers:


ELECTROPHYSIOLOGY

Class Readings/Handouts
(The complete guide is available at www.axon.com/mr_Axon_Guide.html.)

Chapter 1: Bioelectricity
Chapter 2: The Laboratory Setup
Chapter 3: Instrumentation for Measuring Bioelectric Signals from Cells
(Don't get bogged down in the details of Chapter 3)
Chapter 4: Microelectrodes and Micropipettes
Chapter 12: Noise in Electrophysiological Measurements
(Chapter 12 is Supplemental Resource Material)

Student Presentation Options


ELECTROCHEMICAL RECORDING


Exocytosis Measurements (choose one of the following to present; the others can provide context):


Electrochemical Imaging of Drug Transport through Skin (these papers go together as a pair for ONE presentation):
<Juan> H.S. White, "Electrochemical Imaging of Molecular Transport in Skin," The Electrochemical Society Interface, Fall 2003: 30-34.

Multianalyte Microphysiometry (these papers go together as a pair for ONE presentation; the review can help with background/context):

Scanning Electrochemical Microscopy (these papers go together as a pair for ONE presentation; the review can help with background/context):

MASs SPECTROMETRY


Fluorescence; Molecular & Cellular Force Measurements; FTIR Spectroscopy; NMR Spectroscopy & MRI; Mass Spectrometry; Electrochemical Recording; Microfluidics & Cellular Instrumentation; Electrophysiology