Fundamentals of Biomedical Microscopic Imaging

1. **Department** Biomedical Engineering
2. **Number** BME 694 (call number:)
3. **Title of course** Fundamentals of Biomedical Microscopic Imaging
4. **Description** ***(from Course Description Bulletin)***
   Principles and description of microscopy techniques (light, electron and atomic force microscopy) for application to biomedical research.
5. **Level** Graduate and upper level undergraduate
6. **Credits** 3
7. **Class Time Distribution** two times/week for 1.5 h each time
8. **Prerequisites** junior or senior undergraduate or graduate standing
9. **Quarters Offered** Spring
10. **General Information**
    Previous exposure to light microscopy would be helpful.
11. **Exclusions** Limited to senior undergraduates and graduates only
12. **Cross-Listings**
13. **Other Information**
14. **Course Objectives**
    To introduce the physical principles involved in basic light, electron and atomic force microscopy techniques. To understand the strengths and limitations of these microscopic techniques, the sample preparation involved and how they can be selected and/or adapted for specific biomedical applications. To establish fundamental knowledge in proper use of these microscopic techniques.
15. **Textbooks and Other Required Material**
    Recommended books: “Fundamentals of Light Microscopy and Electronic Imaging” by Douglas B. Murphy and “Video Microscopy: The Fundamentals” by Shinya Inoue and Kenneth R. Spring. Other recommended reading include: “Light and Electron Microscopy” by Elizabeth M. Slayter and Henry S. Slayter and “Electron Microscopy: Principles and Techniques for Biologists” by John J. Bozzola and Lonnie D. Russell. The instructor will distribute copies of lecture notes and other relevant reading material as required.

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16. Topics *(including approximate duration)*

Physics of Light and Color- (nature of electromagnetic radiation, definitions of refraction, reflection, diffraction, interference, birefringence, polarization, primary colors, human vision, mirrors, lenses, beamsplitters, laser systems, optical fibers) (**1 lecture**)

Basic Concepts in Microscopy- (resolution, field of view, numerical aperture, image brightness, magnification, depth of field and image formation) (**2 lectures**)

Anatomy of the Microscope- (objective specifications, Köhler illumination, optical aberrations, immersion media, light sources, eyepieces, condensers, and ergonomics) (**1 lecture**)

Sample preparation methods (light microscopy)- (for biomedical applications) (**1 lecture**)

Specialized Microscopy Techniques-1- (differential interference contrast (DIC), phase contrast and dark field microscopy). (**1 lecture**)

Specialized Microscopy Techniques-2 (**Fluorescence microscopy**)- (labeling biomolecules, simple fluoresence microscopy, confocal microscopy, time lapse fluorescence, fluorescence resonance energy transfer (FRET)). (**1 lecture**)

Introduction to Electron Microscopy- (concepts and formulas for light microscopy re-visited for an electron beam, interaction of electron beam with samples, scanning and transmission electron microscopy (SEM and TEM). (**2 lectures**)

Anatomy of an Electron Microscope –(electron optics, lenses and aberrations, column design (SEM vs. TEM), detectors) (**1 lecture**)

Sample Preparation Methods (electron microscopy)- (for biomedical applications) (**1 lecture**)

Specialized Electron Microscopy techniques- (electron diffraction, dark field, energy dispersive spectroscopy (EDX), electron tomography, cryo TEM). (**1 lecture**)

Introduction to Atomic Force Microscopy (AFM)- (principles of AFM, contact and tapping mode, the concept of resolution, contrast, artifacts vs. aberrations, sample preparation methods. (**1 lecture**)

AFM in biomedical imaging- (AFM for real-time and fluid imaging, force measurements and nanolithography. (**1 lecture**)

Advanced microscopic techniques and research applications of microscopy. (2 lectures)

17. Representative Lab Assignments *(if applicable)*

N/A

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18. Grading Plan

Final grade will be based on homework assignments (20%), one mid-term test on theories (20%), a final exam on biomedical applications of microscopy (40%), and a project report on microscopy technique of your choice (20%). The final grade assigned to each student will depend totally upon the performance of the individual student, and will not be affected by the grades of other students. In other words, grades will not be curved.

Biweekly homework sets will involve questions based on microscopy principals discussed in class and visits to "virtual microscopy" sessions available via internet. The mid-term test will be based on understanding the fundamental principals involved in microscopy techniques. The final exam will be geared towards use of microscopic techniques for biomedical applications. In the middle of the quarter, the students will be asked to select a microscopy technique of their choice and compile a report (including references) involving its (a) description and principals, (b) state-of-the art, (c) strengths and limitations and (d) the future.

19. Contribution to Meeting ABET "Professional Component" ***(i.e., to ABET "mathematics and basic sciences, engineering topics, and general education") (if applicable)***

N/A

20. Relationship to ABET-Accredited Program Objectives ***(if applicable)***

N/A

21. Preparation Date 10/25/04

22. Preparer Name Gunjan Agarwal, PhD

23 Contact: Melanie Senitko (BME) : senitko.1@osu.edu

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Lecture Schedule (subject to minor changes):

SP07, BME694, Wed and Fri, at 9:30 to 11:00 am in Bevis 245

1.	What is light, reflection, refraction, lenses and aberrations?	G. Agarwal	Mar 28 (Wed)
2.	Superposition of waves: interference, diffractions and polarization	G. Agarwal	Mar 30 (Fri)
3.	Image formation by a lens	G. Agarwal	Apr 4 (Wed)
4.	Anatomy of a microscope, Koehler alignment, light sources	G. Agarwal	Apr 6 (Fri)
5.	**Light microscopy Lab (parts of a microscope (inverted and upright), Koehler alignment, lens cleaning)**	**G. Agarwal**	**Apr 11 (Wed)**
6.	Fluorescence and Confocal Microscopy	R. Burry	Apr 13 (Fri)
7.	Contrast in Microscopy (dark field, phase contrast, DIC)	G. Agarwal	Apr 18 (Wed)
8.	Sample preparation for light microscopy	R. Burry	Apr 20 (Fri)
9.	**Midterm (objective type, closed book)**		**Apr 25 (Wed)**
10	Introduction to electron microscopy, lenses, beam-sample interactions	G. Agarwal	Apr 27 (Fri)
11	Anatomy of an electron microscope	G. Agarwal	May 2 (Wed)
12	Sample preparation for electron microscopy	Ed Calomeni	May 4 (Fri)
13	Specialized EM techniques	G. Agarwal	May 9 (Wed)
14	**Visit to CMIF (confocal, SEM, TEM)**	**R. Burry**	**May 11 (Fri)**
15	Introduction to atomic force microscopy	G. Agarwal	May 16 (Wed)
16	Biomedical applications of AFM	G. Agarwal	May 18 (Fri)
17	**Visit to AFM Facility**	**G. Agarwal**	**May 23 (Wed)**
18	Advanced microscopy techniques	G. Agarwal	May 25 (Fri)
19	Research applications of microscopy	Anthony Brown	May 30(Wed)
20	**Project report presentations**	**G. Agarwal**	**Jun 1 (Fri)**
	Final Exam (open book)		**Jun 6 (Wed)**