

# Electron Microscopy

BIOL/CBIO 5040/7040  
BIOL/CBIO 5050L/7050L  
Fall 2007

## **COURSE DESCRIPTION:**

This course has both a lecture and laboratory component (3 credit hours each), which are offered every fall semester and are graded separately. Although it is advised to take both concurrently, they may be separated between different years with the stipulation that the lecture portion must be taken before the lab.

The courses cover the theory, fundamental operating principles, and specimen preparation techniques of transmission electron microscope (TEM) and scanning electron microscopy (SEM). The courses are available to graduate students and upper level undergraduates and the lecture portion is co-taught by Drs. Mark Farmer and John Shields. The lab is run by the GLA and supervised by Dr. John Shields.

The EM courses are offered through the Division of Biological Sciences of the UGA (cross listed as VPAT 7040 and CBIO 7040) and cover the use of the instruments, specimen preparation, ultramicrotomy and basic techniques needed to prepare biological and non-biological samples for EM and X-ray microanalyses.

The course syllabus is a general plan for the course; deviations announced to the class by the instructor may be necessary.

## **INSTRUCTORS:**

**Dr. Mark A. Farmer**  
**EM Lab (mornings only)**  
**Room 100 BioSciences Bldg**  
**Phone: (706) 583-8111**  
**Email: [farmer@cb.uga.edu](mailto:farmer@cb.uga.edu)**  
**Office hours are scheduled by appointment.**

**Dr. John P. Shields**  
**EM Lab**  
**Room 151 Barrow Hall**  
**Phone: (706) 542-4080**  
**Email: [jshields@cb.uga.edu](mailto:jshields@cb.uga.edu)**  
**Office hours are scheduled by appointment.**

---

**Yainitza Hernández-Rodríguez (GLA)**  
**EM Lab**  
**Room 154 Barrow Hall**  
**Phone: (706) 542-4080**  
**Email: [yrodriguez@plantbio.uga.edu](mailto:yrodriguez@plantbio.uga.edu)**  
**Office hours are scheduled by appointment.**

**TEXT:**

Bozzola, J.J. and L.D. Russell. 1999. Electron Microscopy: principles and techniques for biologists, 2<sup>nd</sup> Ed. Jones and Bartlett Pub., Sudbury, MA. ISBN 0-7637-0192-0 (hard).

This textbook is also available as an online book at the UGA library.

The lab manual is available online at <http://www.uga.edu/caur/caurmanual.pdf>. Students are responsible for downloading and printing a copy of your own so that you will be able to follow along with the procedures that will be covered in each lab. The first several pages of the lab manual describe in detail the requirements of each lab project, which will be turned in for grades.

**ACADEMIC HONESTY:**

All academic work must meet the standards contained in “A Culture of Honesty.” Students are responsible for informing themselves about those standards before performing any academic work. More detailed information can be found at:

<http://www.uga.edu/ovpi/honesty/acadhon.htm>

**MAKE-UP POLICY:**

Students who miss an exam or a lab with a valid excuse will be allowed to make-up the assignment provided that they contact the instructor within 48 hours of the absence. Pre-arranged valid absences are preferred. Make-up exams may be different from the original exam or may be in an oral format. Assignments due in lab must be turned in within one week of the absence. Failure to hand in make-up assignments or exams by the allotted time will result in no credit for the missed assignment.

**LECTURE:**

Monday, Wednesday, Friday → 11:15 am – 12:05 pm  
Room 216 Biological Sciences

General lecture notes are available online at <http://www.uga.edu/caur/teaching.htm> under either Dr. Farmer’s or Dr. Shields’ PowerPoint presentations/notes depending on who is lecturing on the topic. These may be slightly altered from the versions you will see in class, but if that is the case then we will let you know when the website will be updated with the newer version. It is helpful to download and print the presentations out in handout form (3, 4, or 6 slides per page) prior to lecture so that you can take notes on them. Also on this webpage are links for supplemental detailed information on SEM and TEM lecture background and terms that can prove helpful during studying or for general clarification.

Generally, the class before an exam is set aside for reviewing the materials already covered that could be on that exam. This is structured around students asking questions about topics and can be as long as the class period or shorter if there is a lack of questions. There is no handout or study guide for these review sessions.

### **Assignment of Grades:**

- 2 exams at 100 pts each (200 pts total)
  - Preparation Techniques (100 pts)
  - Physics and Mechanics of EM (100 pts)
- 1 comprehensive final exam at 200 pts
  - Specialized preparation techniques, confocal microscopy, AFM, etc. ( $\geq 100$  pts)
  - Comprehensive material (the remaining points)

**Total = 400 pts**

A	=	400
A-	=	380
B+	=	350
B	=	320
B-	=	315
C+	=	290
C	=	280
C-	=	260
D	=	240

### **Presentation Schedule and Exam Dates:**

**Aug. 17** Introduction & History of EM, Lab Safety (JPS & MAF)

20 Survey of Ultrastructure and Cell Components (JPS)

22 Specimen Preparation (JPS)

24 Fixatives and Buffers (JPS)

27 Preparation Artifacts and Questions (JPS)

29 Dehydration, Embedding Resins, (JPS)

31 Sectioning and Staining (JPS)

#### **Sept. 03 LABOR DAY HOLIDAY**

05 Support Films & Negative Staining (JPS)

07 Critical Point and other Drying Techniques (JPS)

10 Specimen Coating (JPS)

12 Cytochemistry (JPS)

14 Immuno-EM

17 Review Session (JPS)

#### **19 FIRST HOURLY EXAM (100 pts)**

21 Electron Optics – Electron Sources (MAF)

24 Electromagnetic and Electrostatic Lens Theory (MAF)

26 Cryopreservation (JPS)

28 Freeze Fracture (JPS)

#### **Oct. 1 Lens Aberrations (MAF)**

03 Vacuum Systems (MAF)

05 Column Design: SEM vs. TEM (MAF)

08 SEM Design (MAF)

10 Beam and Specimen interactions (JPS)

- 12 Beam-Specimen Interactions: Secondary and Backscattered Electrons (JPS)
- 15 SEM Detectors (MAF)
- 17 Techniques for nanotechnology (TBA)
- 19 Environmental and Variable Pressure SEM (JPS)
- 22 Review Session
- 24 SECOND HOURLY EXAM (100 pts)**

**25-26 FALL BREAK**

- 29 Specialized Preparation Techniques – Material Sci. (TBA)
- 31 High Resolution Electron Microscopy (TBA)
- Nov. 02** Auger Electrons and X-rays (MAF)
- 05 X-rays: EDS vs. WDS (MAF)
- 07 Electron Diffraction (MAF)
- 09 Electron Spectroscopic Imaging and EELS (MAF)
- 12 Imaging and Output Devices (MAF)
- 14 Image Capture – Photography (MAF)
- 16 Digital Imaging (MAF)
- 19 Image Processing and Image Analysis (MAF)
- 20 Tomography (MAF)

**21-23 THANKSGIVING HOLIDAY**

- 26 Confocal Microscopy (JPS)
- 28 Confocal and Multiphoton applications (JPS)
- 30 X-ray Tomography (JPS)
- Dec. 03** Scanning Probe Microscopy (MAF)
- 04 E-Beam Lithography (MAF)
- 05 Review Session

**10 (12:00 – 3:00 pm) FINAL EXAM (200 pts) – Room 216 Biological Sciences**

---

## **LABORATORY:**

Tuesday, Thursday → 2:00 – 4:45 pm

Electron Microscopy Lab\*

Room 155 Barrow Hall

\*also known as the Center for Advanced Ultrastructural Research – CAUR

([www.uga.edu/caur/](http://www.uga.edu/caur/))

Students will be learning many hands-on EM techniques during the laboratory course. Be sure to pay your student fees as laboratory materials for this class will be charged to your student account. You can check your account on OASIS for details.

There will be certain aspects of the lab that will require students to create an image plate and a report (in scientific journal format). Students may turn in rough drafts of plates and/or reports to Dr. Shields or the GLA to ensure that they are correct. It is advised that students have an “Instructions to Authors” handout from a journal that is in their field of study so that they can practice formatting their work to their field’s specifications. Generally the more specific the instructions are, the easier it is for students to format. Students will need to hand the journal instructions to the lab advisors to ensure that they know what the journal expects from its authors. The four projects are as follows, details for each are provided in the lab manual:

**TEM Project** (Mouse liver)

**SEM Project** (Silverberry leaves)

**X-Ray Project** (Unknowns handed out by advisors – undergraduates are not required to create plates or write reports on this topic)

**Individual Project** (This project should be based on your work, but should be designed such that it is feasible to finish and write up the project within the timelines of this course.)

### **Assignment of Grades:**

1. Right to Know (RTK) Signed Sheet ([www.busfin.uga.edu/rtk/index.htm](http://www.busfin.uga.edu/rtk/index.htm)) = 5 pts
2. “Instructions to Authors” (A representative article from the journal with images should be provided) = 5 pts
3. Project Proposal (1-2 pages double-spaced with references in text and literature cited as a separate page) = 30 pts
4. SEM Leaf Project (Plate and Report) = 100 pts
5. Sections on Grids = 10 pts
6. TEM Mouse Project (Plates and Report) = 100 pts
7. X-ray Project (Graduate Students only - Plate and Report) = 50 pts
8. Individual Project (Plates and Report) = 100 pts

Total Points (Graduate Student) = 400 pts

Total Points (Undergraduate Student) = 350 pts

A = 90 – 100%

B = 80 – 89%

C = 70 – 79%

D = 60 – 69%

**Schedule and Deadlines:**

<b>Date</b>	<b>Lab</b>	<b>Lab Topic</b>	<b>Material Due</b>	<b>Pts</b>
Aug 16	01	Intro to CAUR/Lab Safety (RTK)/Intro to digital imaging. (YR & JPS)		
21	02	Specimen Fixation	<b>RTK sheet signed “Instructions to Authors”</b>	<b>5</b>
23	03	Fixation/Dehydration/Embedding		<b>5</b>
28	04	CPD & Stub preparation	<b>Project Proposal</b>	<b>30</b>
30	05	Specimen Coating for SEM		
Sept 04	06	Introduction to SEM		
06	07	SEM Leaf Project (Fixation)		
11	08	Work Day: SEM Leaf Project		
13	09	Digital Image Processing & Photoshop		
18	10	Glass Knife & Block Trimming	<b>SEM Leaf Project</b>	<b>100</b>
20	11	Sectioning		
25	12	Introduction to JEOL 100CX TEM		
27	13	JEOL 100CX Camera Exchange & Development		
Oct 02	14	Staining Grids and JEOL 100CX TEM	<b>Sections on Grids</b>	<b>10</b>
04	15	Check out exams for TEM		
09	16	Support Films (Carbon & Formvar)		
11	17	Negative Staining		
16	18	Specimen Preparation for X-ray & Carbon Coating (JPS)	<b>TEM Mouse Project</b>	<b>100</b>
18	19	Backscatter & X-ray (LEO 982 SEM)		
23	20	X-ray project/ Individual Work day		
25		<b>Fall Break</b> – No Lab		
30	21	X-ray project Day #2	<b>X-ray Project (Grad. Students)</b>	<b>50</b>
Nov 01	22	Electron Diffraction		
06	23	Immuno-EM		
08	24	Immuno-EM Day #2 (scope time)		
13	25	Shadow casts & Replicas (MAF)		
15	26	High Pressure Freezing		
20	27	Cryopreservation & Freeze Fracture	<b>Individual Project</b>	<b>100</b>
22		<b>Thanksgiving Break</b> – No Classes or Lab		
27	28	Project critique	<b>Presentations</b>	<b>50</b>
29	29	<b>Presentations</b>		
Dec 10		Final Revised Projects handed in by 5:30pm	<b>All Final Projects</b>	<b>-</b>