

# **BIBC 103: Biochemical Techniques**

## **Fall 2010**

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858-822-2032

**Office hours:** Mondays 1-2 pm

**Lecture:** Monday/Wednesday/Friday 10:00 am to 10:50 am, HSS 1330

**Labs:** York 2310 and 2332

Tuesday/Thursday: 11:00 am – 3:00 pm

Wednesday/Friday: 11:00 am – 3:00 pm

### **Course Objectives:**

This course will introduce some of the experimental techniques used in biochemistry and molecular biology. These include methods for purifying proteins, expressing recombinant proteins in bacterial cells, and analyzing biological molecules by electrophoresis, Western blotting, and enzyme activity assays. The laboratory work will consist of three multi-day projects, as well as some smaller, single-day experiments. Through the laboratory work we will try to develop the skills required to design and interpret the data from scientific experiments. This scientific reasoning is central to work done in any field of science. In keeping with this, the importance of good experimental design, including the use of appropriate controls, will be highlighted in all experiments. Also, as this is an introductory lab course, all lab work will emphasize the learning of basic lab skills (including dilutions, good pipetting technique, and basic statistical analysis) and good lab practices (such as good notebook keeping).

### **Materials Required:**

1. Biochemical Techniques Lab Manual (available from the Bookstore)
2. Bound laboratory notebook
3. Safety glasses
4. Lab coat

**Course Requirements and Grading:** Your final grade for the class will be calculated using the following criteria:

Exams (3 at 160 pts. each)	480 points
Lab Reports (2 at 220 pts. each)	440 points
Data Write-up (mini report)	40 points
Lab Notebook Checks (4 at 5 pts. each)	20 points
Pre-Lab Pop Quizzes (4 at 5 pts. each)	20 points
<b>Total</b>	<b>1000 points</b>

**Point Cutoffs for Grade Assignments:** (Cutoffs may be lowered at the instructor's discretion.)

910-1000 = A	790-799 = C+
900-909 = A-	705-789 = C
890-899 = B+	695-704 = C-
810-889 = B	600-694 = D
800-809 = B-	0-589 = F

### **Course Web Site:**

Many of the course materials are available only through the course website on WebCT (<https://webctweb.ucsd.edu>). All students will need to be able to access this site. Once you are enrolled in the class you will have access to the site using your ACS username and password. Be sure to check the course website frequently for announcements and updates on assignments. Items such as lab report guidelines and image files of gels and other data will be provided through the website. The 'Additional Materials' folder contains additional background material for some of the experiments. Use the Discussion Board to ask questions on material from lecture or lab. The instructor will check the Discussion Board daily to answer questions, but students are encouraged to answer questions as well. This is a handy resource for last minute questions that come up during late night studying for an exam.

### **Lab Notebooks:**

You will be expected to keep a formal laboratory notebook for all of the work you do in lab. The notebook should be bound (spiral bound or composition book style are both okay), and should have numbered pages with a table of contents (it is okay to write these in). You will need to hand in either photocopies or carbon copies of your notebook pages for the experiments that are written up as lab reports. Notebook entries should be in chronological order, with each project or set of experiments together and easily referenced by the table of contents. Each page should have a brief title for the experiment and the date on which the work was performed. Refer to the course lab manual for some other tips on entering information into your notebook. Starting on the second day of lab, you will need to have the following entered in your notebook at the beginning of each lab session:

**A. From the previous day's experiment:** all of your data entered in labeled spaces, and any analysis for that experiment completed. Analysis includes any calculations and graphs that may be required to analyze the data. Your TA will tell you what analysis needs to be done for each experiment. There should also be a brief summary (not more than a few sentences) of the experiment that states how well the procedure worked and

any major conclusions from the data.

**B. For the current day's experiment,** a brief purpose explaining what you are doing that day (one or two sentences is fine), and appropriately labeled spaces and tables in which you will enter any data collected that day. Also, make sure that your table of contents and page numbering is up to date.

There will be four unannounced notebook checks, worth 5 points each, where your TA will inspect your lab notebook. The TA will most likely choose certain labs or analysis to focus on at each check.

**Lab Manual and Pop Quizzes:** It is important to carefully read the pertinent sections of the lab manual before coming to class. The experiments will proceed much more smoothly, and you will get a lot more out of them, if you have read through the procedure and understand what you are doing. To encourage you to read the lab manual before class, there will be four unannounced quizzes that will be given at the beginning of lab.

**Lab Attendance Policies:**

Attendance at each lab session is mandatory. The first unexcused absence will result in 25 points being deducted. The second unexcused absence will result in an F for the course. If you know that you need to miss a lab session, discuss this with the instructor (not the TA, they are not authorized to give you permission) to see if it will be possible to make up the lab session or excuse you from the lab with no consequences. Please bring this to the instructor's attention as soon as you know that it will be an issue. **Only the instructor can excuse an absence. Two unexcused absences will result in the student failing the course.**

**Turning in Lab Reports:**

We will be using the website Turnitin.com for lab reports in addition to turning in hard copies to your TA. Lab reports submitted to the Turnitin.com site do not need to have graphs, tables, or attachments, but you may include them if it is easier. Lab reports must be submitted to Turnitin.com before midnight of the due date, and a hard copy of the report (including all text, plus all tables, graphs, attachments, or anything else called for in the lab report guidelines) must be given to your TA at the beginning of the due date lab session. Lab reports not turned in at the beginning of the lab session, or not submitted to Turnitin.com by the end of the day will be considered one-day late. Ten points will be deducted for each working day that the lab reports are late (hard copy and Turnitin.com). Students agree that by taking this course all required papers would be subject to review for textual similarity by Turnitin.com for the detection of plagiarism. All submitted papers will be included as source documents in the Turnitin.com reference database solely for the purpose of detecting plagiarism of such papers. Use of the Turnitin.com service is subject to the terms of use agreement posted on the Turnitin.com site.

**Make-up Exams:**

Please note that it is extremely burdensome for the instructor and TAs to have to prepare and proctor make-up exams. Missing a scheduled exam will only be excused for medical reasons where documentation can be provided. At the instructor's discretion, a missed exam that is excused will either be dropped from the student's point total for the class, or

made up by an oral exam scheduled within one week of the original exam.

	Dates	Experiment/Activity	Lab Manual Chapter
<b>Wk 1</b>	Sept. 28, 29	Introduction to Micropipettes	Lab 1
	Sept. 30 Oct. 1	Making a pH buffer; Quantitative Measurements	Lab 2
<b>Wk 2</b>	Oct. 5, 6	LDH 1: Initial purification of LDH from crude homogenate: centrifugation, ammonium sulfate precipitations; prepare size exclusion column	Lab 3
	Oct. 7, 8	LDH 2: Affinity chromatography purification of LDH	Lab 4
<b>Wk 3</b>	Oct. 12, 13	LDH 3: Size exclusion chromatography purification of LDH	Lab 5
	Oct. 14, 15	LDH 4: Activity assays; Bradford protein assays	Lab 6
<b>Wk 4</b>	Oct. 19, 20	LDH 5: Native gel electrophoresis of LDH with activity stain	Lab 7
	Oct. 21, 22	Bioinformatics: LDH and Fluorescent Proteins <b>Exam 1 in lecture</b>	Lab 18
<b>Wk 5</b>	Oct. 26, 27	Fluorescent proteins (FP) 1: Plasmid preps <b>Lab report 1 due</b>	Lab 12
	Oct. 28, 29	FP 2: Restriction enzyme digest; Agarose gel electrophoresis	Lab 13
<b>Wk 6</b>	Nov. 2, 3	FP 3: Make competent cells and transform with plasmid	Lab 14
	Nov. 4, 5	FP 4: Purification and analysis of fluorescent proteins	Lab 15
<b>Wk 7</b>	Nov. 9, 10	FP 5: SDS-PAGE of fluorescent proteins;	Lab 16
	Nov. 11, 12	Holiday—No lab	
<b>Wk 8</b>	Nov. 16, 17	FP 6: Examine SDS-PAGE gels; Set up lysozyme crystallization <b>Exam 2 in lecture</b>	Lab 17 Lab 19
	Nov. 18, 19	MAP kinase Western blot (MAPK): Sea urchin fertilization	Lab 8
<b>Wk 9</b>	Nov. 23, 24	MAPK: SDS-PAGE and electroblotting <b>Lab report 2 due</b>	Lab 9
	Nov. 25, 26	Holiday—No lab	
<b>Wk 10</b>	Nov. 30 Dec. 1	MAPK: Immunodetection and analysis of X-ray films; Examine lysozyme crystals	Lab 10 Lab 19
	Dec. 2, 3	<b>Exam 3 in lab</b>	