

BIBC 103: Biochemical Techniques

Winter Quarter, 2020

Instructor: Michael Burg, Ph.D.
Office: H&SS 1145LB
mburg@ucsd.edu

Office Hours: Mon 12:30pm-1:30pm (if requested) (Also available during lab; other office hours can be arranged as needed)

Lecture: Center 214 Tue/Thu 8:00– 9:20 AM

Labs: All labs Tue/Thu. B01:9:30AM-1:20PM:York 3306; B02:9:30AM-1:20PM:York 3406
B03:2:30PM-6:20PM:York 3306 B04:2:30PM-6:20PM:York 3406

This course will introduce some of the experimental methods used in biochemistry and molecular biology, with an emphasis on those techniques used to study proteins. You will gain a conceptual understanding of, and some hands-on experience in, various protein purification techniques, expression and purification of recombinant proteins from bacterial cells, and methods for analyzing the different properties of proteins. The laboratory work will consist of three multi-day projects, as well as some smaller, single-day experiments. As this is an introductory lab course, all lab work will emphasize the learning of basic lab skills and good lab practices.

More importantly, this course is designed to give an appreciation of what science is and how it works. Science is not just a bunch of random facts...it is a process! It is easier to understand biology, or any field, when you understand why we know what we know about it. Understanding how information in biology is brought to light is just as important as the information itself. Through the laboratory projects we will develop the skills necessary to interpret data from experiments in order to answer questions about biological systems, and to design experiments to answer new questions. In keeping with this, the importance of good experimental design, including the use of appropriate controls, will be highlighted in all experiments. A complete list of the learning goals and expected outcomes for the course can be found on Ted.

Materials Required:

- 1) Biochemical Techniques Lab Manual, 2019/2020 Edition (available from the Bookstore)
- 2) **Bound** laboratory notebook (not loose leaf; do not need carbon copies)
- 3) Safety glasses
- 4) Lab coat

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

Exams (120 pts. and 260 pts.)	380 points
LDH purification table analysis	145 points
Sea Urchin Lab Report	255 points
Crystallization writeup	30 points
Lab Notebook (3 X 30 pt notebook checks) (LDH,Sea Urchin,FP)	90 points
Lab Quizzes (2 at 35 pts. each)	70 points
Bioinformatics	30 points

Total	1000 points
--------------	--------------------

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

990-1000	A+	790-799	C+
900-989	A	700-789	C
890-899	B+	600-699	D
800-889	B	<600	F

In addition: Extra credit pts are available

Greater than 80% CAPE response: 6 pts

Course Web Site:

Many of the course materials are available only through the course website on TritonLink Education, or Ted (<https://ted.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.

Lab Notebooks:

You will keep a formal laboratory notebook for a lot of your work in the class. A well-kept lab notebook serves as a portfolio of the experiments and techniques you have performed, something that can be useful when interviewing for research internships and laboratory jobs. Your notebook needs to be bound (no loose pages), but composition books are okay. The notebook does not need to have carbon copy pages, you will not have to turn in copies of notebook pages. See page *ix – x* in the lab manual for how to format your notebook and what information it should contain. Pay particular attention to the following:

- Write the **experiment date** in the upper left-hand corner of **each page**. Make all entries in chronological order. You do not need page numbers or a table of contents—you will index your entries by the experiment date.
- Project title** following the date on each page (*e.g.*, LDH Purification and Analysis). Be sure to separate the three projects in your notebook.
- Brief introduction stating overall purpose/overview of lab (see example lab report)**
- Experiment title** underneath the project title on each page. This should be a single sentence indicating the specific procedure that was performed.
- Briefly list any changes to the procedures from the lab manual. Other than that, you do not need to write out procedures.
- Raw data and important observations: Enter numerical values in an organized table. For large numbers of numerical values collected electronically, you may paste printer tapes or a printout of the Excel spreadsheet into the notebook. These must be permanently fixed; you will not get

credit for items loosely tucked into the pages. Also include any important observations (be brief). Look for prompts in the lab manual for what to include.

- g. Data analysis: Include any calculations, statistics, or graphs immediately following the raw data. This should be done for any and all data you collect (with the exception of the exercises in Lab 1). Graphs and plots should be done using Excel (or another graphing package) and should be labeled in text. They need to be printed and pasted into your notebook. Be sure they look professional!—ask for help with graphing in Excel if you are having trouble.
- h. All electrophoresis gel and Western blot images should clearly labeled with text, printed, and pasted into your notebook.
- i. Include a brief statement of the conclusions from the experiment. This may be a single sentence to simply verify that you successfully concluded that procedure on days where you don't collect any data, to a short paragraph describing the results of a multi-day experiment. You should also succinctly describe anything that went wrong with that experiment. What would you do differently if you had to do the experiment again?
- j. Your lab notebook should not contain lecture notes!

Your notebook should be kept up to date as you carry out each lab. Analysis (including plots and gel images) must be completed and added to the notebook by the lab period following collection of the data. Your IA will perform unannounced lab notebook checks throughout the quarter.

Lab Quizzes and Exams:

The purpose of the lab quizzes is to address the following: Are you keeping up with the material? Are you prepared for that day's lab? Have you been attending lecture? The quiz dates are given in the lab schedule. They will begin precisely at the scheduled lab start time (so be ready to go when you come in) and will take 30 minutes. They can cover any material from the weeks of class prior to that quiz, but will focus on understanding the purpose of the lab projects and how each experiment fits into this, the basic concepts underlying the procedures, and simple mathematical and analytical skills based on what you have actually done in lab.

The two exams are cumulative and will be problem solving-based. They may include some basic questions on the concepts we have covered, but will emphasize taking the information you have learned and extrapolating to solve problems you have not seen before. Practice questions will be given on Ted to help you prepare for the exams.

Lab Attendance Policies:

Attendance at each lab session is mandatory. An unexcused absence will result in 10 points being deducted. If you know that you need to miss a lab session, discuss this with the instructor (not the IA, 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:

Lab reports are due at the beginning of lab on due date listed in the lab schedule. In addition to the hard copy turned in to your lab IA, an electronic copy of the report must also be submitted to Turnitin.com, which is accessed through Ted. The report must be submitted to Turnitin.com before the hard copy is turned in, and the hard copy must contain the Turnitin.com submission receipt in the appendix. Lab reports not turned in at the beginning of the lab session on the due date will be considered one-day late. Ten points will be deducted for each working day that the lab reports (five points for LDH write-up) are late. Students agree that by taking this course all required papers will 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.

Re-Grade Policy on Lab Reports: Your lab reports will be graded by your IA, based on the same lab report guidelines (general and specific) that you receive. Note that I work closely with all the IAs to ensure that the grading is accurate and equivalent between sections. If you disagree with the grading of your lab report, discuss this with your IA first to get clarification on why points were deducted.

Making Up Quizzes and Exams:

Please note that it is extremely burdensome for the instructor and IAs to have to prepare and proctor make-up exams. If you disagree with the grading of your exams, discuss this with your IA first to get clarification on why points were deducted. Missing a scheduled quiz or exam will only be excused for medical reasons where documentation can be provided.

LECTURES WILL CORRESPOND TO LAB TOPICS AND

BIBC103 Winter 2020 SCHEDULE

Lab Schedule

	Dates	Experiment/Activity	Lab Manual Chapter
Wk 0	Sept. 26		
Wk 1	Jan 7	Safety orientation and waste streams, introduction to equipment, pipetting and dilution skills	Lab 1
	Jan 9	Elrctrophoresis & SDS-PAGE	Lab 2
Wk 2	Jan 14	LDH 1: Initial purification of LDH from crude homogenate: centrifugation, ammonium sulfate precipitations; prepare size exclusion column	Lab 3
	Jan 16	LDH 2: Affinity chromatography	Lab 4
Wk 3	Jan 21	LDH 3: Size exclusion chromatography Quiz 1	Lab 5
	Jan 23	LDH 4: Activity assays; Bradford protein assays	Lab 6
Wk 4	Jan 28	LDH 5: SDS-PAGE of LDH purification fractions	Lab 8
	Jan 30	LDH 6: Native gel electrophoresis of LDH with activity stain; E Lysozyme crystallization 1: Set up hanging drops round 1 examine SDS PAGE Gel	Lab 7 Lab 20
Wk 5	Feb 4	Exam 1 during lecture	
	Feb 4	Sea Urchin fertilization; prepare cell lysates	Lab 9A
	Feb 6	MAPK Western blot—SDS PAGE and electroblotting	Lab 10
Wk 6	Feb 11	MAPK Western blot—Immunodetection; LDH Purification Table Analysis Due	Lab 11
	Feb 13	ELISA for phospholipase C activity; Examine crystals round 1	Lab 12

			Lab 20
Wk 7	Feb 18	Work up ELISA data, create figures for lab report Crystallization 2: Set up hanging drops round 2	Lab 12 Lab 20
	Feb 20	Bioinformatics 1: Investigation of an unknown melanoma gene Quiz 2 in lab	Lab19 A
Wk 8	Feb 25	Bioinformatics 2: Modeling protein structures	Lab 19 B-D
	Feb 27	FP (fluorescent proteins); Make competent cells and transform with plasmid	Lab 13 parts A & B; Lab 15
Wk 9	Mar 3	Purification and analysis of fluorescent proteins Sea Urchin report due	Lab 16
	Mar 5	SDS-PAGE of fluorescent proteins; Examine crystals 2	Lab 17 Lab 20
Week 10	Mar 10	Examine SDS-PAGE gels	Lab 18
	Mar 13	Exam 2 in lab	