BIBC 103: Biochemical Techniques

Winter Quarter, 2012

Instructor: Goran Bozinovic, Ph.D.

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Office Hours: In lab or by appointment

Lecture: CENTR 113: Tue, Thu 12:30-1:50 AM

Labs: First floor Bonner Hall; Tue / Thu 2:30-6:30 PM Wed / Fri 9 AM – 1 PM

Section	Lab Room	TA Email	
C01	BH 1309	Sreekanth Kumandan	sreekumandan@gmail.com
C02	BH 1329	Smith, Taylaur	t9smith@ucsd.edu
C03	BH 1413	Penrose, Harrison	hpenrose@ucsd.edu
C04	BH 1309	Rajakumar, Nimesh	nrajakum@ucsd.edu
C05	BH 1329	Schulman, Andrew	aschulma@ucsd.edu
C06	BH 1413	Eric Soule	esoule@ucsd.edu

Support Staff/Lab Safety Officer: Joe Stagg (Bonner Hall 1402)

Telephone: 534-2195; email: jstagg@ucsd.edu

Course Objectives:

This course will introduce various laboratory techniques used in biochemistry and molecular biology. These will 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. 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 in lab everyday:

- 1) Biochemical Techniques Lab Manual (available from the Bookstore)
- 2) Bound laboratory notebook with 100 numbered duplicate pages
- 3) Pen (Lab notes must be in ink)
- 4) Calculator
- 5) Safety glasses
- 6) Lab coat
- 7) Proper Attire (no open toed shoes)

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

Exams (3 at 240 pts. each)	720 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
Total	1240 points

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

1140-1240	Α	966-991	C+
1115-1139	A-	893-965	С
1090-1114	B+	856-892	C-
1016-1089	В	744-855	D
992-1015	B-	0-743	F

Course Web Site:

Many of the course materials are available only through the course website on 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. Use the Discussion Board to ask questions on material from lecture or lab. The instructor and TAs 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 acceptable), and should have numbered pages with a table of contents (it is okay to write these in). Lab notes must be written in ink. 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 <u>previous day's experiment</u>: 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 <u>current day's experiment</u>, 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 why you are doing them. 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. If you are more than 10 minutes late to lab, or you leave lab before your group is done, you will be counted as absent for the day. An unexcused absence will result in 10 points being deducted from the associated lab report. 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.**Also, everyone is expected to be an active participant in every experimental procedure. Failure to make a meaningful contribution towards completing the laboratory experiment/activity will

result in points being deducted from the laboratory report score.

Turning in Lab Reports:

We will be using the "Turnitin" via TED for lab reports in addition to turning in hard copies to your TA. Lab reports submitted to the TED / Turnitin site do not need to have graphs, tables, or attachments, but you may include them if it is easier. Lab reports must be submitted 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 <u>beginning</u> of the due date lab session. Lab reports not turned in at the beginning of the lab session, or not submitted to TED 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 will be subject to review for textual similarity by Turnitin 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 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.

Lab Schedule:

	Dates	Experiment/Activity	Lab Manual
			<u>Chapter</u>
Wk 1	Jan 10/11	Organization/Safety; Introduction to Micropipettes	Lab 1
	Jan 12/13	Making a pH buffer; Quantitative Measurements	Lab 2
Wk 2	Jan 17/18	LDH 1: Initial purification of LDH from crude homogenate: centrifugation,	
		ammonium sulfate precipitations; prepare size exclusion column	Lab 3
	Jan 19/20	LDH 2: Affinity chromatography purification of LDH	Lab 4
Wk 3	Jan 24/25	LDH 3: Size exclusion chromatography purification of LDH	Lab 5
	July 26/27	LDH 4: Activity assays; Bradford protein assays	Lab 6
Wk 4	Jan 31/ Feb 1	LDH 5: Native gel electrophoresis of LDH with activity stain	Lab 7
	Feb 2/3	MAP kinase Western blot (MAPK): Sea urchin fertilization	Lab 8
		Exam 1 in lecture, Thursday, Feb 2nd	
Wk 5	Feb 7/8	MAPK: SDS-PAGE and electroblotting Lab Report 1 due	Lab 9
	Feb 9/10	MAPK: Immunodetection	Lab 10
Wk 6	Feb 14/15	MAPK: IP ₁ ELISA to detect phospholipase C activation; Examine Western blot films	Lab 11
	Feb 16/17	Bioinformatics: LDH and Fluorescent Proteins	Lab 18
Wk 7	Feb 21/22	Fluorescent proteins (FP) 1: Plasmid preps Exam 2 in lecture, Tuesday, Feb 21st	Lab 12
	Feb 23/24	FP 2: Restriction enzyme digest; agarose gel electrophoresis	Lab 13
Wk 8	Feb 28/29	FP 3: Make competent cells and transform with plasmid;	Lab 14
		Set up lysozyme crystallization	Lab 19
	Mar 1/2	In-class Review Session: Lab - TBA	Lab 15
Wk 9	Mar 6/7	FP 4: Purification and analysis of fluorescent proteins Lab report 2 due	
	Mar 8/9	FP 5: SDS-PAGE of fluorescent proteins	Lab 16
Wk 10	Mar 13/14	FP 6: Examine SDS-PAGE gels; Examine lysozyme crystals	Lab 17
			Lab 19
	Mar 15/16	Exam 3 in lecture – Thursday, March 15	
		FP data write up due (mini-report)	

Academic Integrity

Cheating will not be tolerated. The administrative policy on Academic Dishonesty outlined by UCSD will be followed. Students caught cheating during an exam or quiz will be removed and given a "zero" for that exam or quiz. A report will also be filed with the Academic Integrity Coordinator. Cheating includes (but is not limited to) plagiarism, talking during tests, or making use of forbidden materials during the test. Students are permitted to bring only non-programmable calculators and writing implements to exams. Tampering with graded exams will result in a failing grade for that exam.

During laboratory sessions, student cooperation and collaboration is highly encouraged. This includes discussion of experimental data with fellow students during lab hours. After the laboratory session is over, however, you are required to work on your own. *Each student must hand in an independently written and independently thought-out data analysis for each lab*. If you are caught working with another student on your lab report, both of you will receive a "zero" for that lab report, and you might be reported to the Academic Integrity Coordinator.

It is NOT acceptable to use any old lab reports to assist you in any way. If you happen to be in possession of old copies of lab reports for this class, it is best that you do not even look at them, since they could unintentionally influence the way that you write your own report. If we discover that you have used an old lab report in any way, you will automatically receive a "zero" for that lab report, and you might be reported to the Academic Integrity Coordinator. While your lab reports will be returned to you, you are NOT permitted to share them with anyone for any reason. If we find that you have shared your lab report with anyone, you will be reported to the Academic Integrity Coordinator, even if you have already completed the class. You are required to read, understand, and sign the "BIBC103 Integrity Policy" which governs the manner in which the academic work in this class is completed.