BIMM 101 Recombinant DNA Techniques

Fall 2013

Dr. Goran Bozinovic

gbozinovic@ucsd.edu

HSS 1145G

Office hours: by appointment

TAs: Gutierrez, Josue Rafael jrg004@ucsd.edu partlo, William N. wpartlo@ucsd.edu

Lectures: Mon, Wed, Fri 8 – 8:50 AM SEQ 147

Labs: Mon, Wed 9 - 1 York 4318, 4332

Course Website: http:// ted.ucsd.edu

- Lectures will cover the theory behind the experiments performed in lab. The quizzes and exam will have questions on the material that will be covered during lectures.

Learning objectives:

- Learn the theory behind molecular techniques, and the applications of the methodologies in biological research
- Become proficient at basic molecular biology techniques
- Learn the importance of proper controls in designing experiments and interpreting results
- Improve lab math skills and ability to graph data correctly
- Learn to make logical conclusions from experimental data
- Become familiar with bioinformatics databases and applications
- Learn to find, read, and evaluate primary literature
- Become aware of the implications of the technology for society

Required texts:

BIMM 101 Lab Manual from University Readers From Genes to Genomes by Dale (1st or 2cd edition) on reserve at BML and electronic version available from UCSD computer http://onlinelibrary.wiley.com/book/10.1002/0470856912 Readings on TED (ted.ucsd.edu)

Required Materials - needed by second day of class:

Labcoat (the bookstore has cheap ones)
UV blocking safety glasses (also at bookstore)
Lab notebook with carbon copies (bookstore or Grove general store)
Fine point Sharpie for labeling – get a dark color
A calculator

Remember that lab attendance is required – if you miss two labs, you will be asked to drop the course. If you are ill, you must leave a message with your instructor, not your TA, and make up the lab in a way that we will determine. You must be on time for lab; the TAs go over the experiments at the beginning of lab, and also quizzes are administered then.

Course Requirements

- **1. Lab Notebook**: It is mandatory that you keep a complete lab notebook. The notebook must contain everything that you did in the lab, including:
- Any changes in the protocol
- All data/results
- All calculations done during experiments
- Observations
- **2. Take home assignments:** there will be two take-home assignments
 - dilutions 10 pts Due on Oct 7th
 - DNA quantification 20 pts, <u>Due on Oct 21st</u>
- **3. Quizzes**: There will be 4 scheduled quizzes (dates TBD) during lab periods. Each quiz is worth 10 points. They will be given at the beginning of lab, and collected 15 minutes later. If you arrive after the quiz has been handed out, you will not be able to make it up.
- **4. Lab Reports**: 4 lab reports throughout the quarter: first two are worth 60 points each, and last two are worth 40 points each. While you will be collecting and sharing data with a lab partner, and you are welcome to discuss your results with your classmates, you must hand in your own lab report, written in your own words. You will be penalized for copying another lab report or for handing in the same (or very similar, such as just a few words changed here and there) lab reports as your partner.

All lab reports should include:

- 1. Purpose of the experiment: this section should be BRIEF no more than a few sentences: simply state why you are doing the experiment.
- 2. Results should include the following:
- data or data analysis
- figures, gels (or representations thereof)
- any sample calculations
- a brief statement about what each result means
- 3. Discussion:
- Note any unusual observations
- Discuss success or failure of the experiment if there was a problem, discuss probable source.

Lab reports should have no more than 5 pages of text – figures can go on separate pages. More specific description of the lab report can be found in the course TED page. Although the lab report will be submitted electronically, all carbons from the labs associated with a lab report must be handed in class the day the lab is due.

Lab report - Late policy.

Lab reports are due at the beginning of lecture on the assigned due date (must be electronically submitted before 8 AM of the due date).

Penalty for turning lab reports late:

- 5 points if handed in later on the same date;

After the first late day, you lose 2 points/day, so

- -7 points if handed in anytime the next day
- -9 points if handed in the 3rd day etc.

Lab report due dates:

Labs to be included in the Lab Report	Due date
Labs 3-8A,	Oct 30
Labs 9-15A, excluding 14c	Nov 25
Labs 16- 18A	Dec 9
Labs 18B, 19	Dec 9

- ** Please submit your Lab Reports at the beginning of the lab. Lab reports 3 and 4 (submit them stapled together) should be submitted on the morning of the final exam on Monday, Dec 9th. You may also submit your reports to your TAs earlier.
- **5. Lab attendance**: Attendance is taken within the first 15 minutes of every lecture session. If you are ill, please notify me (gbozinovic@ucsd.edu).
- **6. Lab performance:** There are no points for lab performance per se. However, your effort, attitude, and the success of your experiments will be considered when assigning the final grade, especially if you are on the borderline between two grades. If you miss one lab with no excuse, you will lose 5% from your final grade. If you miss two labs, you will receive an F for the course.
- **7. Exams**: There will be two exams **Midterm** (100 pts) on <u>Wednesday</u>, <u>Nov 13 at 8 AM IN THE LAB</u>, and **Final exam** (150 pts) on <u>Monday</u>, <u>December 9th. at 9 AM IN THE LAB</u>. Final exam is cumulative. Depending on the performance of the class, the exam scores might be adjusted.
- The exam adjustment policy:
- 1) If any student receives a 100% on the exam it will NOT be curved;
- 2) If any student receives a score between 95 and 100%, that will be the new maximum score (for example if the highest grade is 96, everyone's score will increase by 4 points);
- 3) If the highest score is less that 95% then that student's score will be the new 95% (for example if the highest grade is 89, everyone's score will increase

by 6 points).

Final Exam

Grading:

Dilution assignment 10 pts Lab 2 (DNA quantification) assignment 20 pts 4 Quizzes (10 points each) 40 pts

4 Lab reports (1,2 - 60 points each

3,4 - 40 pts200 pts Midterm Exam 100 150 pts

Total possible points: 520

Please make sure you regularly check your scores in TED to make sure no errors have occurred.

Letter grades will be assigned as follows:

Grade Overall class percentage

A+, A, A- 98, 92, 90

B+, B, B- 88, 82, 80

C+, C, C- 78, 72, 70

D+, D, D- 68, 62, 60

F Below 60

Note: Just coming to lab does not ensure that you will get a passing grade in the class. You must hand in all assignments and get passing scores on those assignments (an average of 70) to get a C- in the class.

Policy on cheating: Anyone caught cheating (which includes but it is not limited to plagiarizing lab reports, cheating on a test or guiz, or changing an answer for a regrade) will be reported to the Academic Integrity Office.

BIMM 101 Fall 2013 Student contract:

1. I understand that if I am late for lab on allowed to take the quiz and will receive a	
Name	Date
All lab reports for the class must be in work in your own words. While discussion encouraged, each student on their own mustigures, graphs, and tables. The submission contain shared work is forbidden, and will reboth reports. The exception to this is when supplied to each member of the group (spec photographs). In this case the labeling of the independently. If you have questions about your work with others and unauthorized collar or T.A. for clarification. Because lab reports are to be your own to copy to any extent current or past laborated other students. This is known as plagiarism student to present the work of others as his/cheating on an exam. Directly copying mate putting it in your own words is also plagiarism reference. Plagiarism in lab reports is rigored Students are required to upload an electronic Turnitin.com, where the report is screened were ports in the Turnitin database. All incident turned in to the Academic Integrity Coordinate Integrity of Scholarship (www-senate.ucsd.estudents found to have committed plagiarism receive both an administrative (decided by the penalty (decided by the instructor). Furtherm retained in the Turnitin database. Similarity also reveal the name of the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the student who proone's own lab report to other students to allowed the	ndependently written, i.e., your own of data among lab partners is tomplete all text, references, of reports by lab partners that sult in points being deducted from a figure is the raw data that is diffically absorption spectra and geleat figure must be done the difference between discussing aboration, please ask your instructor with work in your own words, you may story reports that were written by which is a direct attempt by the her own, and is no different than erial from other sources without m, even if the source is cited as a pusly sought out and penalized. It is of plagiarism checker against all is of plagiarism will automatically be tor. Following UCSD's Policy on du/manual/appendices/app2.htm), or or other academic misconduct will the Council of Deans) and academic more, all submitted reports are hits by the plagiarized material. Giving ow them to copy material from that be pursued and penalized as
2. I understand that if I plagiarize a lab re Turnitin.com, the matter will go to the Ac	ademic Integrity Office on
campus. I also understand that if I give a the lab in a subsequent quarter, and he o will also be subject to disciplining by the	r she plagiarizes my lab report, l
Name	 Date

Tentative Schedule

<u>Date</u>		
9. 30	Lab 1	A. PipettingB. DilutionsC. Bacteriological Techniques
10. 2	Lab 2	A. Agarose Gel Electrophoresis
10. 7	Lab 3	 A. Introduction to bioluminescence B. Course Objectives C. P1: Isolation of chromosomal DNA from <i>Vibrio fischeri</i>
10. 9	Lab 4	A. P2: Isolation of chromosomal DNA from <i>Vibrio</i> fischeri
10. 14	Lab 5	 A. Spectrophotometric Analysis of Vibrio DNA B. Sal I restriction digestion of Vibrio fischeri and pGEM DNA
10. 16	Lab 6	 A. Agarose gel electrophoresis of Sall restriction digest and estimate of DNA quantities from gel B. Ligation of <i>Vibrio fischeri</i> DNA with pGEM DNA C. Bacteriological techniques: streaking fir single colonies on agar plates
10. 21	Lab 7	 A. Ligation assessment by gel electrophoresis B. Transformation of competent E. coli DH5α cells
10. 23	Lab 8	A. Count colonies and re-streak glowing colonies from original transformation
10. 28	Lab 9	 A. Amplification of <i>lux</i>AB gene by PCR B. Start overnights for experiment to size and seq. <i>V. fischeri Sal</i> I inserts C. Bioinformatics computer Lab 1
10. 30	Lab 10	 A. PCR success assessment by gel electrophoresis B. Alkaline lysis miniprep: purification of plasmid DNA C. Restriction Digest with Xbal and EcoRi D. Clean up luxAB PCR Products

11. 4	Lab 11	 A. Clean up Xbal and EcoRI digest of pGEM B. Quantification of digests from gel C. Ligation of pGEM and luxAB inserts D. Bioinformatics Lab 2: Analysis of non-glowing colony inserts
11. 6	Lab 12	 A. Making competent DH5α cells B. Transformation of ligation products into DH5α cells C. Biodiversity: insect DNA and PCR of the CO1 gene
11. 11	Holiday – Veterans' Day	
11. 13	Lab 13	 A. Screening for colonies containing <i>lux</i>AB by adding exogenous aldehyde B. Run gel to check CO1 PCR C. Clean up PCR products and send for sequencing
11. 18	Lab 14	A. Analyze barcode resultsB. Isolate cheek cell DNA and set up PCR for PTC
11. 20	Lab 15	A. PTC taste test and PCR product digestB. Run Gel to PTC PCR product and analyze results
11. 25	Lab 16	A. Set up C. elegans plates for RNAi experiment
11. 27	Lab 17	A. Observe worm phenotypes and isolate RNAB. Quantitate RNA and set up quantitative RTPCR
12. 2	Lab 18	A. Analyze results of RTPCR measurements of unc-22 mRNA
12. 4	Lab 19	Review

FINAL EXAM: Monday, Dec 9th 9 – 11 AM in LAB