

**BIMM 101      Recombinant DNA Techniques      Summer I 2016**

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HSS 1145G

Office hours: during the lab or by appointment

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**Lectures:**      Tue-Fri 9:30-10:50      York 4080A

**Labs:**            Tue-Fri 11-3                      York 4318 / 4332

**Course Website:** <http://triton.ed.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 (1<sup>st</sup> or 2<sup>nd</sup> edition) on reserve at BML and electronic version available from UCSD computer

<http://onlinelibrary.wiley.com/book/10.1002/0470856912>

Readings on tritonEd ([triton.ed.ucsd.edu](http://triton.ed.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 IA, and make up the lab in a way that we will determine. You must be on time for lab; the IAs go over the experiments at the beginning of lab, and also quizzes are administered then.

**!!! Mandatory Lab Safety Training Prior to Lab Participation:**

All students, enrolled or waitlisted, for the laboratory courses below are REQUIRED to demonstrate an understanding of general lab safety and UCSD Undergraduate Biology lab rules and policies. Completing the on-line Lab Safety Training and passing the Lab Safety Assessment fulfills this requirement.

Enrolled students who miss the first lab meeting, or do not pass the Lab Safety Assessment, will have their lab space revoked. It is the students responsibility to drop the course or risk receiving a "W" or non-passing grade in the course.

- The Lab Safety Training and Assessment are available at any time, but the assessment will apply to a specific quarter.
- The assessment must be completed and passed BEFORE the first lab meeting. Plan ahead!
- The self-study training and assessment should take approximately 1-hour to complete.
- The assessment portion is timed. Students will have a maximum of 30-minutes to complete the assessment.
- Students may take the assessment as many times as it takes to pass.
- Students will be certified once the assessment is passed. Certification information will be available to lab instructors for verification prior to the first class meeting. Certification is valid for one quarter.

**<http://biology.ucsd.edu/education/undergrad/course/ug-labs.html>**

## 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

- There will be three random notebook checks starting the second week of class in order to maintain GLP notebook keeping. Each check is worth 5 pts.

**2. Take home assignments:** there will be two take-home assignments

HW #1: Dilutions and DNA Quantification- 30 pts – due Tue, July 5

HW #2: Statistical Analysis (Lab 10) – 30 pts – due Friday, July 15

**3. Quizzes:** There will be 4 scheduled quizzes during lab periods starting July 5, each 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:** Three lab reports are to be submitted throughout the quarter: lab report 1 is worth 100 points, and LR2 / LR3 are worth 50 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. Specific guidelines for each lab report are posted on the course website.

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 TritonEd 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 11 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:**

<u>Labs to be included in the Lab Report</u>	<u>Due date</u>
LR1 - Labs: 4-11	July 19
LR2 - Labs 14-15	July 29
LR3 - Labs 16-18	July 29

\*\* Please submit your Lab Reports at the beginning of the lab. Lab reports 2 and 3 (submit them stapled together) should be submitted on the morning of the final exam on Friday, July 29. You may also submit your reports to your IAs 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. Exam:** There will be two exams – Midterm (100 pts) on Tuesday, July 19<sup>th</sup>, and Final exam (150 pts) on Friday, July 29<sup>th</sup>. 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 than 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 pts).

**Grading:**

HW #1	30
HW#2	20
Notebook Checks (3)	15
4 Quizzes (10 points each)	40
LR 1	100
LR2 + LR 3	80
Midterm Exam	100
Final Exam	150

**Total possible points: 535**

Please make sure you regularly check your scores in TritonEd 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 quiz, or changing an answer for a regrade) will be reported to the Academic Integrity Office.

**BIMM 101 Summer 2016 Student contract:**

**1. I understand that if I am late for lab on a day a quiz is given, I will not be allowed to take the quiz and will receive a 0 score for that quiz.**

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Name Date

All lab reports for the class must be independently written, i.e., **your own work in your own words**. While discussion of data among lab partners is encouraged, each student on their own must complete all text, references, figures, graphs, and tables. The submission of reports by lab partners that contain shared work is forbidden, and will result in points being deducted from both reports. The exception to this is when a figure is the raw data that is supplied to each member of the group (specifically absorption spectra and gel photographs). In this case the labeling of that figure must be done independently. If you have questions about the difference between discussing your work with others and unauthorized collaboration, please ask your instructor or I.A. for clarification.

Because lab reports are to be your own work in your own words, you may not copy to any extent current or past laboratory reports that were written by other students. This is known as plagiarism, which is a direct attempt by the student to present the work of others as his/her own, and is no different than cheating on an exam. Directly copying material from other sources without putting it in your own words is also plagiarism, even if the source is cited as a reference. Plagiarism in lab reports is rigorously sought out and penalized. Students are required to upload an electronic version of each lab report to Turnitin.com, where the report is screened with a plagiarism checker against all reports in the Turnitin database. All incidents of plagiarism will automatically be turned in to the Academic Integrity Coordinator. Following UCSD's Policy on Integrity of Scholarship ([www-senate.ucsd.edu/manual/appendices/app2.htm](http://www-senate.ucsd.edu/manual/appendices/app2.htm)), students found to have committed plagiarism or other academic misconduct will receive both an administrative (decided by the Council of Deans) and academic penalty (decided by the instructor). Furthermore, all submitted reports are retained in the Turnitin database. Similarity hits by the plagiarism checker will also reveal the name of the student who provided the plagiarized material. Giving one's own lab report to other students to allow them to copy material from that report is also academic dishonesty, and will be pursued and penalized as rigorously as for the student committing the plagiarism.

**2. I understand that if I plagiarize a lab report and it is detected by tritonEd.com, the matter will go to the Academic Integrity Office on campus. I also understand that if I give a lab report to a student who takes the lab in a subsequent quarter, and he or she plagiarizes my lab report, I will also be subject to disciplining by the Academic Integrity Office.**

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Name Date

## Tentative Schedule, Summer 1, 2016

Week	Class	LAB
1	Tue, 6.28	<b>LAB 1</b> A. Pipetting B. Dilutions C. Calibration of a pipette D. Mol. Bio. Review
	Wed, 6.29	<b>LAB 2</b> A. Agarose gel electrophoresis on two DNA samples of unknown size and concentration (estimating using standard curve)
	Thu 6.30	<b>LAB 3 *Computer Lab*:</b> A. Image J analysis of gel electrophoresis results & graphing
	Fri 7.1	<b>LAB 4</b> A. Introduction to bioluminescence B. Cloning the <i>luxA</i> and <i>luxB</i> gene into <i>E.coli</i> : outline of experiments C. Part 1: Isolation of chromosomal DNA from <i>Vibrio fischeri</i>
2	Tue 7.5	<b>LAB 5</b> A. Finish purification of chromosomal DNA from <i>Vibrio fischeri</i> D. <b>Computer Lab</b> - Bioinformatics Part I: exploring the Lux operon on NCBI + primer design HW # 1 Due – Dilutions and DNA Quantification
	Wed 7.6	<b>LAB 6</b> A. Spectrophotometric analysis of <i>Vibrio</i> DNA B. Design and set up experiment to vary conditions of PCR (amplifying <i>V. fischeri luxAB</i> genes)
	Thu 7.7	<b>LAB 7</b> A. Checking the success of the PCR reaction by gel electrophoresis B. <b>Computer Lab:</b> Using Image J to analyze PCR results + make graph C. Time to repeat PCR if needed or expand upon variations from Lab 6
	Fri 7.8	<b>Lab 8</b> A. Run gel of repeats (if necessary) B. Clean up best <i>luxAB</i> PCR product from lab 6 C. Restriction digest of <i>luxAB</i> PCR products and pGEM with <i>XbaI</i> and <i>EcoRI</i> D. <b>Computer Lab:</b> Bioinformatics Part II and III (restriction digestion) & Part IV Primer Design
3	Tue 7.12	<b>LAB 9</b> A. Clean up <i>XbaI</i> and <i>EcoRI</i> digest of pGEM B. Quantification of digests from gel C. Ligation of pGEM and <i>luxAB</i> inserts

	<b>Wed</b> <b>7.13</b>	<b>Lab 10</b> A. Transformation of competent cells with ligation products B. Practice statistical analysis: ANOVA and post-hoc test
	<b>Thu</b> <b>7.14</b>	<b>Lab 11</b> A. Screening for clones containing <i>luxA</i> by adding exogenous aldehyde (Assessing ligation efficiency (blue/white colony counting) → Pool data from whole class to do statistical analysis of results). B. Plan Synthetic Biology project C. Start overnights of cultures containing plasmids with different promoters
	<b>Fri</b> <b>7.15</b>	<b>Lab 12</b> A. Alkaline lysis miniprep: purification of plasmid DNA from overnight cultures B. Setting up digests of Biobrick plasmids <b>HW #2 due – Statistical Analysis Due</b>
<b>4</b>	<b>Tue</b> <b>7.19</b>	<b>Lab 13</b> A. Removing the stuffer fragment from the plasmids containing the promoter sequences B. Gel purification of the DNA fragment containing the RFP sequence Ligating plasmids with promoter sequences and RFP sequence <b>Midterm Exam in Lab; LR 1 due</b>
	<b>Wed</b> <b>7.20</b>	<b>Lab 14</b> A. Transformation of competent cells with RFP ligation products B. Begin PTC: Do PTC taste test, isolate cheek cells DNA and set up PCR
	<b>Thu</b> <b>7.21</b>	<b>Lab 15</b> A. Analyze effect of promoters on RFP expression (fluorometer measurements). Pool class data for analysis. B. Pick one plasmid and set up overnights (will isolate and send for sequencing to confirm promoter-RFP ligation and reading frame) B. Digest PTC cheek cell PCR and run gel
	<b>Fri</b> <b>7.22</b>	<b>Lab 16</b> A. Begin RNAi project: Set up <i>C. elegans</i> plates B. Isolate plasmid and run gel to estimate concentration, send for sequencing
<b>5</b>	<b>Tue</b> <b>7.26</b>	<b>Lab 17</b> A. Observe worm phenotypes and isolate RNA B. Quantitate RNA and set up quantitative RTqPCR C. <b>Computer lab:</b> Analyze plasmid sequencing results (supp. hand-out)
	<b>Wed</b> <b>7.27</b>	<b>LAB 18</b> A. Computer Lab: Analyze results of RT-qPCR measurement of <i>unc-22</i> mRNA
	<b>Thu</b> <b>7.28</b>	<b>LAB – Review/Paper discussion</b>
	<b>Fri</b> <b>7.29</b>	<b>Final EXAM in Lab</b> <b>LR 2 and LR 3 due in lab</b>



