BIMM 101 Recombinant DNA Techniques | Winter 2016

Sections B01 and B02

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Lecture: Tuesday, Thursday at 12:30-1:50pm CSB 005

Laboratory: Tuesday and Thursday, at 2:30 - 6:30 pm in York 4318 (B01) or York 4332 (B02)

Office Hours: TBD

Required materials

1. BIMM 101 Lab Manual from **Soft Reserves** (available on campus at the Soft Reserves office)
2. Other readings occasionally posted on TED
3. i>clicker, registered on TED
4. Lab Coat (must be to knees)
5. UV-blocking safety glasses
6. Long pants or equivalent, close-toed and closed-heel shoes
7. Fine point Sharpie (dark color) for labeling
8. Carbon copy or carbonless copy notebook (bookstore) for taking lab notes
9. Calculator (cell phones are not allowed during quizzes)

Calendar: Please see the condensed calendar at the bottom of the syllabus. A more detailed calendar is found separately on TED. Note that this calendar is subject to change - although I aim to stick to the posted calendar it is possible we will deviate from it. If the schedule changes, I will let you know.

Learning goals:

- Apply knowledge of the theory behind molecular techniques, and the applications of the methodologies in biological research, to explain experimental steps and troubleshoot results
- Demonstrate proficiency at basic molecular biology techniques
- Explain the importance of proper controls in designing experiments and interpreting results
- Perform basic lab math skills, statistical analysis, and graphing and graphing
- Draw logical conclusions from experimental data and justify conclusions
- Use basic bioinformatics databases and applications
- Learn to find, read, and evaluate primary literature

Learning in this course

This course is designed to be a collaborative environment for everyone to learn together and construct a shared understanding of the material. Active participation both in class and in discussion sections is expected. Being able to communicate understanding, and confusion, is critical to success in any discipline, and is very useful for learning[1]. To encourage collaboration, class activities and discussions will be done in groups, and grades will not be assigned on a curve.

I like to use class time to work on applying our knowledge, troubleshooting difficult topics, and practice solving problems. Hence, there are targeted readings and quizzes set up that are due before class. This way, we all come to class prepared to contribute to valuable discussions.

Instead of memorization, we will focus on developing an understanding of fundamental concept. Therefore, quizzes will include questions that are based on solving problems in new contexts.

1 Smith et al., 2009. http://www.sciencemag.org/content/323/5910/122.short

Grading
There are three components of grading in this course: Participation, Lab Mini Reports, and Quizzes

Participation: 20%

a. **Pre-lab/lecture quizzes on TED, 5%**
   Quizzes are due before every class. Instructions on what to read before taking the quiz are posted on TED for each quiz. These quizzes are meant to test low-level knowledge of the content and lab procedures. You get two attempts at the quiz, and your final score is the higher of the two attempts. Your lowest score is dropped from your total score.
b. **Lab notebooks, 10% (10 randomly graded, 1% each)**
   We will hand out instructions on what to put in your lab book during the first lab.
c. **Clicker participation (not for correctness), 3%**
   A note about clickers: you can purchase an iClicker2 at the bookstore. iClicker 1 has had issues with "remembering" class settings even within the course of a lecture (you can use iClicker 1, but please be aware of these issues). If you participate in 90% of clicker questions in class, you will get full points. Because you only need 90% participation for full points, if you forget your clicker one day do not worry about it.
   ****Please bring your clicker on day 1****

d. **Professionalism in the lab (2%):** Working in a lab means you will be interacting with many other people: peers, your instructional assistant, and the professor. It is important to use professional and respectful behaviour in the lab, both here and if you pursue lab work opportunities outside of the course. This mark is based on your overall professional behaviour in the lab, as judged by your IA, professor, and peer group. 0 = multiple instances of unprofessional behaviour, 1 = few (e.g. 1 or 2) instances of unprofessional behaviour, 3 = no instances of unprofessional behaviour. What is unprofessional behaviour? Let's discuss this at the start of the lab course.

Laboratory mini reports and assignments: 25%
Guidelines and rubrics for each of the mini reports and assignments will be posted on TED and due dates are on the calendar (below) and on TED. Reports will be submitted to Turnitin on TED and hard-copies must be submitted in person within 10 minutes of the start of your lab.
There are 5 mini reports and a barcoding assignment:

Gel electrophoresis – 2%
PCR variations – 5%
LuxAB – 5%
Synthetic Bio – 5%
RNAi – 6%
Sequence or PTC analysis – 2%

Quizzes: 55%
Starting in Week 2, there will be short quizzes at the start of every Tuesday class, on the material covered the week prior. There will be 9 quizzes, your top 8 scores will be used → 8 x 4.375% each = 35%
The final quiz, during the last lab, is cumulative and worth 20%.
Quizzes are open book (lab manual + class notes) no electronic devices.

Absences: Lab attendance is required – if you miss one lab with no excuse, you will lose 5% from your final grade. 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. Two late arrivals to lab will be counted as one absence.

Grades will be based on your percentage in the course:

97+ = A+	80 up to 82 = B-
94 up to 97 = A	76 up to 79 = C+
90 up to 93= A-	72 up to 75 = C
87 up to 89 = B+	67 up to 71= C-
83 up to 86 = B	0 up to 66= D
	Below 60 = F

This course is not graded on a curve (i.e. 20% of students getting A, B, C, and such), and the ability to do well in the course is not dependent on others doing poorly. You must have an average passing score on assignments and quizzes in order to pass the course (an average of 60 on your assignments and an average of 60 on the quizzes).

Laboratory safety
Safety precautions are crucial in the laboratory setting. As such, appropriate personal protective equipment (PPE), including laboratory coats that cover to the knees, UV-blocking safety glasses or googles, long pants or equivalent, and closed-toe and closed-heel shoes, are required. **You must take the lab safety module quiz prior to the start of Lab 2**. You can find the safety module here: http://biology.ucsd.edu/education/undergrad/course/ug-labs.html

Academic integrity (https://students.ucsd.edu/academics/academic-integrity/index.html)
Integrity of scholarship is essential for an academic community. The University expects that both faculty and students will honor this principle and in so doing protect the validity of University intellectual work. For students, this means that all academic work will be done by the individual(s) to whom it is assigned, without unauthorized aid of any kind. Anyone caught cheating (includes plagiarizing lab reports, cheating on a test, or changing an answer for a re-grade) will be reported to the Academic Integrity Office.

Late and missed assignments and quizzes
Late assignments will be subject to a 10% deduction per day (note that assignments handed in after the first 10 minutes of lab are considered late) up to a maximum of 2 days late (after which you will receive a 0). There are no make-up quizzes offered except in the case of a documented medical or family emergency (in which case the instructor will decided how to go about the make-up testing).

Inclusion and accessibility (http://disabilities.ucsd.edu)
Any student with a disability is welcome to contact us early in the quarter to work out reasonable accommodations to support your success in this course. Students requesting accommodations for this course due to a disability must provide a current Authorization for Accommodation (AFA) letter issued by the Office for Students with Disabilities (OSD), which is located in University Center 202 behind Center Hall. Students are required to present their AFA letters to faculty and to the OSD Liaison in the Division of Biological Sciences in advance so that accommodations may be arranged. For further information, contact the OSD at 858-534-4382 or osd@ucsd.edu.

Letters of recommendation: Lab courses can offer an opportunity for students and instructors to become better acquainted compared to large lecture classes. Often, students from lab courses will

request letters of recommendation for program applications, etc. I am happy to write letters of recommendation for deserving students, but I have some criteria about writing letters. I like to write letters that can offer more than what is on your transcript, so I look for: participation in class and lab, demonstrations of critical thinking, leadership, professionalism, evidence a student takes responsibility for their learning, maturity, making contributions to discussions. If you ask me for a letter of recommendation, before deciding if I will write one, I may ask that we have a conversation about these criteria, and I may ask you to explain to me when and how you think you have demonstrated these behaviours. Also I recommend that you keep your graded assignments, because if you ask me for a letter I will want to see them.

Course Calendar

For computer lab days it is much easier if you bring your own lap top

Week	Date	Lecture	Lab	Assignments Reading = quiz on TED
1	**Tues** **Jan 5**	Intro Dilutions	**LAB 1** A. Pipetting B. Dilutions C. Calibration of a pipettemen D. Mol. Bio. Review	
	Thurs **Jan 7**	Agarose gel electrophoresis (AGE) & Continue gene structure review	**LAB 2** A. Agarose gel electrophoresis on two DNA samples of unknown size and concentration B. Describing results and drawing conclusions (computer lab/on TED)	Reading due
2	**Tues** **Jan 12**	Quiz #1 on Week 1 p-value & t-test Figures & results description (writing)	**LAB 3 *Computer Lab** A. Computer Lab: Image J on Lab 2 AGE pictures B. Computer Lab: Graphing of Image J results (and intro to basics of graphing) Computer Lab: introduction to basic statistics (t-test) to compare two means	AGE mini report due at the start of Lab 5
	Thurs **Jan 14**	LuxAB operon Cloning Part I DNA extraction	**Lab 4** A. Introduction to bioluminescence B. Cloning the *lux*A and *lux*B gene into *E.coli*: outline of experiments C. Part 1: Isolation of chromosomal DNA from *Vibrio fischeri*	Reading due

3	**Tues Jan 19**	Quiz #2 on Week 2 PCR, experimental design, bacteriological techniques	**LAB 5** A. Finish purification of chromosomal DNA from *Vibrio fischeri* B. Practice bacteriological techniques D. Design PCR experiment	Reading due AGE mini report due at the start of Lab
	Thurs Jan 21	Lab 6 content + TBD	**LAB 6** A. Spectrophotometric analysis of *Vibrio* DNA B. Set up experiment to vary conditions of PCR (amplifying *V. fischeri luxAB* genes) C. Record results from bacterial plates	Reading due
4	**Tues Jan 26**	Quiz #3 on Week 3 TBD	**LAB 7** A. Checking the success of the PCR reaction by gel electrophoresis B. **Computer Lab:** Using Image J to analyze PCR results C. Time to repeat PCR if needed or expand upon variations from Lab 6	
	Thurs Jan 28	Digests & ligation, more cloning, mini report info	**Lab 8 *Computer Lab – BRING YOUR OWN COMPUTER** A. Run gel of repeats (if necessary) B. Clean up best *lux*AB PCR product from lab 6 C. Restriction digest of *lux*AB PCR products and pGEM with *Xba*I and *Eco*RI D. **Computer Lab:** Bioinformatics 1 (NCBI to identify operon features)	Reading due PCR mini report due Lab
5	**Tues Feb 2**	Quiz #4 on Week 4 Mini report info	**LAB 9** A. Clean up *Xba*I and *Eco*RI digest of pGEM B. Quantification of digests from gel C. Ligation of pGEM and *lux*AB inserts B. **Computer Lab:** Bioinformatics 2: primer design (instructions provided on TED)	Reading due

	Thurs Feb 4	Transforming cells, more on stats	**Lab 10** A. Transformation of competent cells with ligation products B. **Computer Lab:** Instructions on TED. Intro to and doing basic stats analysis with some supplied ligation data (or other data)	Reading due PCR mini report due at start of lab
6	**Tues Feb 9**	Quiz #5 on Week 5 Synthetic biology	**Lab 11** A. Screening for clones containing *lux*A 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	Reading due
	Thurs Feb 11	Continue synthetic biology, plasmid prep, ANOVA	**Lab 12 *Computer – bring your own** A. Alkaline lysis miniprep: purification of plasmid DNA from overnight cultures B. Setting up digests of Biobrick plasmids C Computer Lab: ANOVA and post-hoc analysis to analyze pooled ligation data (from Lab 11)	Reading due Ligation efficiency mini report due Lab 14
7	**Tues Feb 16**	Quiz #6 on Week 5 or paper (TBD) TBD	**Lab 13** A. Removing the stuffer fragment from the plasmids containing the promoter sequences B. Gel purification of the DNA fragment containing the RFP sequence C. Ligating plasmids with promoter sequences and RFP sequence	Reading due
	Thurs Feb 18	SNPs and RFLPs	**Lab 14** A. Transformation of competent cells with RFP ligation products B. Begin PTC: Do PTC taste test, isolate cheek cells DNA and set up PCR	Reading due Ligation efficiency mini report due at start of Lab

8	**Tues Feb 23**	Quiz #7 on Week 7 DNA sequencing & continue SNPS/RFLPs	**Lab 15** 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	Synthetic bio mini report due Lab 17
	Thurs Feb 25	*C elegans*, RNAi & intro RT-PCR	**Lab 16** A. Begin RNAi project: Set up *C. elegans* plates B. Isolate plasmid and run gel to estimate concentration, send for sequencing	Reading due
9	**Tues March 1**	Quiz #8 on Week 8 Finish RT-PCR and do qPCR Mini report info	**Lab 17 *Computer Lab** A. Observe worm phenotypes and isolate RNA B. Quantitate RNA and set up quantitative RTqPCR C. Computer lab: Analyze plasmid sequencing results (supplemental hand-out)	Reading due Synthetic bio mini report due start of lab
	Thurs March 3	CRISPR/Cas-9 mediated genomic editing Mini report info	**Lab 18 *Bring computer** A. Analyze results of RT-qPCR measurement of *unc*-22 mRNA	RNAi mini report due March 10
10	**Tues March 8**	Quiz #9 on Paper (take-home questions) Paper Discussion	**Lab – Review**	
	Thurs March 10	No class *open office hours*	**Final Quiz during lab time**	RNAi mini report due