BIMM 101 Recombinant DNA Techniques - Fall 2016
Sections F01, F02, F03 and F04

Instructor: Chris Day cdday@ucsd.edu Office: HSS 1145LA

Instructional Assistants:

F01	Angela Nicholson;	a5nichol@ucsd.edu
F02	Laura Chipman;	lchipman@ucsd.edu
F03	JD Gregerson;	jcgreger@ucsd.edu
F04	Matt Mitchell;	mbmitche@ucsd.edu

Lecture: Tuesday, Thursday 12:30-1:50pm SOLIS 104

Laboratory: Wednesday, Friday, 9am-12:50pm in York 4318 (F01) or York 4332 (F02)
Wednesday, Friday, 2pm-5:50pm in York 4318 (F03) or York 4332 (F04)

Office Hours: Tuesday and Thursday 9-10 am *we often have time in lab or at the end of lab when they end early (which is often), so please take advantage of these times to discuss things with me too.

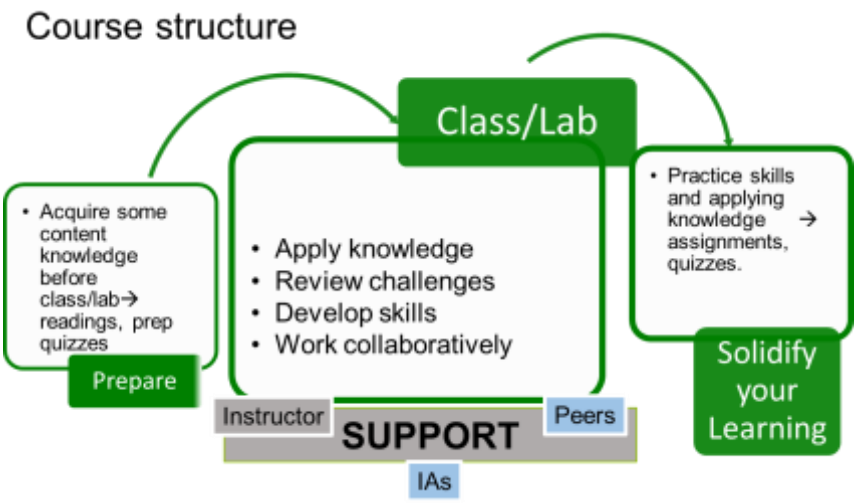
Required materials

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 tubes
8. Carbon copy or carbonless copy notebook (bookstore) for taking lab notes
9. Calculator or cell phone calculator

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
- Apply knowledge of molecular biology concepts relevant to our work to explain 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
- 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 lab 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 communication and collaboration, we will frequently use class time to work on problems in groups.

We like to use class time to work on applying knowledge, troubleshooting difficult topics, and practice solving problems. Hence, it is expected that you will prepare before coming to class, reviewing basic background information about the lab and/or relevant content. This will be encouraged through targeted readings and in-class quizzes. The more prepared you are for class and lab, the more fruitful our discussions can be.

Instead of memorization, we will focus on developing an understanding of fundamental concepts and as they apply to the experiments. Therefore, tests will include questions that are based on solving problems in new contexts or data interpretation and not necessarily on memorizing facts.

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. **Lab notebooks, 10% (10 randomly graded, 1% each)**
   Instructions about what to include in your notes will be posted on TED.

b. **Clicker participation (not for correctness), 5%**
   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.

c. **Lab efficiency and professionalism (5%):** It is important to be diligent when working in the lab: make sure you are following protocols, pay attention to supplies, and use your time effectively. It is also very important to work collaboratively and effectively with others, including dividing tasks equally (one person should not do all tasks). Your lab efficiency and professionalism score will be based on two components:
    i. For efficiency and effectiveness. This is not to say that mistakes are not permitted, mistakes happen. However, if you *chronically* make mistakes, misuse supplies, perform unsuccessful work, you will be docked points.
    ii. For professionalism and collaboration. This mark is based on observations of your behaviour in the lab.

Laboratory mini reports and assignments: 30%

Guidelines and rubrics for each of the mini reports and assignments will be posted on TED and due dates announced on TED and in class. Reports will be submitted to Turnitin on TED and hard-copies must be submitted in person within 5 minutes of the due date time.

There are 5 mini reports and an assignment:

Gel electrophoresis mini report– 3%
PCR variations mini report – 5%
Ligation efficiency – 6%
Synthetic Bio – 7%
RNAi – 9%

Quizzes and Final: 50%

Starting in Week 1, there will be a short quizzes at the start of Wednesday lab. This will be on material covered the prior week and on upcoming material (this should encourage you to read ahead!). There will be 8 quizzes, your top 7 scores will be used → 7 x 4% each = 28%.

The final quiz, during the last lab, is cumulative and worth 22%. Quizzes will be 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 get in touch with me, 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+	94 up to 97 = A	90 up to 93= A-
87 up to 89 = B+	83 up to 86 = B	80 up to 82 = B-
76 up to 79 = C+	72 up to 75 = C	67 up to 71= C-
60 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.

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 decide 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.

Course Calendar

Week	Date	Class or Lab	Lecture
0	**Thursday Sept 22 & Friday Sept 23**	**LAB 1** A. Pipetting B. Dilutions C. Calibration of a pipettemen D. Mol. Bio. Review	Intro
1	**Tues Sept 27 & Wed Sept 28**	**LAB 2** **Quiz 1** A. Agarose gel electrophoresis on two DNA samples of unknown size and concentration (estimating using standard curve)	Agarose gel electrophoresis
	Thurs Sept 29 & Friday Sept 30	**LAB 3 *Computer Lab*** A. Image analysis of gel electrophoresis results & graphing	Graph/data interpretation? Or start on PCR
2	**Tues Oct 4 & Wed Oct 5**	**Lab 4** **Quiz 2** A. Part 1 & 2: Isolation & purification of chromosomal DNA from *Vibrio fischeri* B.Spectrophotometric analysis of *Vibrio* DNA	Introduction to luxAB opeon & bioluminescence DNA extraction

	Thurs Oct 6 & Friday Oct 7	**LAB 5** **Agarose Gel electrophoresis mini report due at start of lab** A. **Computer Lab -** Bioinformatics Part I: exploring the Lux operon on NCBI + primer design B. Plan PCR experiment	PCR and experimental design (controls)
3	**Tues Oct 11 & Wed Oct 12**	**LAB 6** **Quiz 3** A. Set up PCRs (amplifying *V. fischeri luxAB* genes)	More on PCR
	Thurs Oct 13 & Friday Oct 14	**Lab 7 *Computers** A. Checking the success of the PCR reaction by gel electrophoresis B. **Computer Lab:** Using Image J to analyze PCR results + make graph C. Time to repeat PCRs	Paper disucssion
4	**Tues Oct 18 & Wed Oct 19**	**Lab 8 *Computers** **Quiz 4** A. Run gel of repeats (if necessary) B. Clean up best *lux*AB PCR product from lab 6	Cloning and restriction digestion

		C. Restriction digest of *lux*AB PCR products and pGEM with *Xba*I and *Eco*RI D. **Computer Lab:** Bioinformatics Part II and III (restriction digestion) & Part IV Primer Design	
	Thurs Oct 20 & Friday Oct 21 **PCR variations mini report due**	**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	Paper discussion
5	**Tues Oct 25 & Wed Oct 26**	**Lab 10** **Quiz 5** A. Transformation of competent cells with ligation products B. Plan promoter mutants project (synthetic biology) C. Start overnights of cultures containing plasmids with different promoters	Transformation, intro to promoter mutants project
	Thurs Oct 27 & Friday Oct 28	**Lab 11** A. Counting blue/white colonies & screening for clones containing *luxAB* by adding exogenous aldehyde B. Pool data from whole class to do statistical analysis of results (ligation efficiency).	More on syn bio project or paper

		C. Alkaline lysis miniprep: purification of plasmid DNA from overnight cultures (promoter mutants project)	
6	**Tues Nov 1 & Wed Nov 2**	**Lab 12 *Computers** **Quiz 6** A. Setting up digests of Biobrick plasmids B. Statistical analysis of ligation data plus working on report	 Data analysis (ANOVA) Review ligation efficiency report expectations
	Thurs Nov 3 & Friday Nov 4 **Ligation efficiency mini report due**	**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	 Review of what's happening in lab + start paper
7	**Tues Nov 8 & Wed Nov 9**	**Lab 14** **Quiz 7** A. Transformation of competent cells with RFP ligation products	 Paper discussion
	Friday Nov 11 - Veterans Day Holiday. No lab or class on Thursday and Friday.		

8	**Tues Nov 15 & Wed Nov 16**	**Lab 15 *Computers**	
		A. Analyze effect of promoters on RFP expression (fluorometer measurements). Pool class data for analysis.	Predicting effects of promoter mutants - looking at data from journal articles
		B. Statistical analysis of results	
		C. Optional: choose RFP colony to grow up and send for sequencing	
		C. Begin RNAi project	
	Thurs Nov 17 & Fri Nov 18	**Lab 16**	
	Promoter-RFP (synthetic biology) mini report due		
		A. Observe worm phenotypes and isolate RNA	RT & qPCR
		B. Quantitate RNA and set up quantitative RTqPCR	
		C. Optional: purify plasmid, run gel to check concentration and send for sequencing	
9	**Tues Nov 22 & Wed Nov 23**	**Lab 17 *Computers** **Quiz 8**	
		A. Computer Lab: Analyze results of RT-qPCR measurement of *unc*-22 mRNA	Planning data analysis
		B. Optional: analyze sequencing results	
		*Lab clean-up	
	Thursday 24 - Thanksgiving Holiday. No lab or class on Thursday and Friday.		
10	**Tues Nov 29 & Wed Nov 30**	Paper discussion & review	Paper discussion

	Thurs Dec 1 & Friday Dec 2	**Final Quiz during lab time**	Review
	Monday Dec 5th	**RNAi mini report due**	