TECHNIQUES IN MOLECULAR BIOLOGY SYLLABUS

Biol 319 – Fall 2012

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Required Lab Manual: Caporale, D. 2012. Techniques in Molecular Biology: A Laboratory Manual. Department of Biology, UW -Stevens Point. For purchase in Biology Main Office (TNR 167).

Required 3-Ringed Binder: Purchase a small 3-ringed binder to place your lab manual in, as well as lined paper for note taking and assignments.

Class Meetings:
Section 1: Tues 10:00am – 12:50pm; Sci B212
Section 2: Thur 10:00am – 12:50pm; Sci B212

Course Objectives: 1) to understand basic molecular approaches to answer a variety of biological questions, and 2) to gain experience in designing experiments & using molecular laboratory techniques.

Grading:

Assignment	Lab Reports % pt
Lab 1: Gel electrophoresis	**10**
Lab 2a: 1st RAPD Fingerprinting **Lab 2b: 2nd RAPD Fingerprinting**	**30**
Lab 3a: DNA isolation & quantification **Lab 3b: PCR: Detection of *Borrelia*** **Lab 3c: PCR: Detection of *Anaplasma*** **Lab 3d: DNA sequencing**	**40**
Protocol Order / Preparedness / Participation	**20**

Grading Scale: The grading scale above is firm. There will be no borderline grades. If you attend and participate in class, read your manual and formulate a protocol order **prior** to lab time, and **ask questions**, then you'll find success in accumulating points. Guidelines on how to write a lab report are in the lab manual.
Late protocol orders are not accepted. Late lab reports are subject to 10% off each day late.

Attendance Policy: If you miss a lab, you hurt yourself and your partner's grade. It is extremely difficult to make up a lab. Students who must miss a lab due to religious observances or participation in university sanctioned events should notify me within the first 3 weeks of the beginning of class, so makeup arrangements can be made. The only other valid excuses for missing a lab or deadline are death in the family, violent illness, or accident. In such cases: (1) you must provide evidence of some kind (e.g. note from health center), **and** (2) you must reschedule **within 24 hr** after the date of the deadline.

Academic Misconduct: You are responsible for the honest completion and representation of your work and for the respect of others' academic endeavors. Any act of cheating, plagiarism, or academic misconduct is subject to the penalties outlined in UWS Chapter 14. For more info, visit:
http://www.uwsp.edu/centers/rights/RRBOOKLET8-2005-06.pdf

TECHNIQUES IN MOLECULAR BIOLOGY SCHEDULE

Week	Topic	Lab Investigation & Pages	Assignment Due
1	Intro, Prepare 1X TAE buffer Pour Gel	iii 1: 1-4	
2	Gel Electrophoresis: run gel & analyze Design RAPDs Project	1: 5-7 2: 8-11	PO, Collect tissue
3	DNA Isolation & Quantification of species of your choice	3a: 18-20 with revisions	PO
4	Intro to RAPD Technique & Analysis RAPD PCR with 1st primer, Pour 1st gel	2: 8-13	PO, Gel Electrophoresis Report Due
5	Flagging for Ticks in Schmeeckle Reserve		
6	Run RAPD gel, pour 2nd gel RAPD PCR of 2nd primer	2: 8-14	PO
7	Run 2nd RAPD gel, Isolate Tick DNA	2: 13-14 3a: 17-19	PO
8	Quantify Tick DNA, PCR for *Borrelia*, Pour a gel Create 0/1 matrix & tree from 1st RAPD image	3a: 20 3b: 26-30 2: 14-16	PO
9	Run *Borrelia* PCR gel, PCR for *Anaplasma*, Create 0/1 matrix & tree from 2nd RAPD image	3b: 28-30 3c: 30 2: 15-16	PO
10	Nested PCR for *Anaplasma*, Pour gel Combine 0/1 matrices and Construct a tree	3c: 31, 28 2: 15-16	PO
11	Run *Anaplasma* gel, Purify pos. *Borrelia* & pos. *Anaplasma* PCR products, Cycle-sequence	3c: 28-30, 32 3d: 33-37 3d: 38-39	PO, RAPDs Report Due
12	**Thanksgiving Break!!!!**		
13	Purify cycle-sequence products, Prepare for sequencing	3d: 39-40 3e: 41-45	
14	Edit sequences, Perform a Blast Search to Identify Species	3f: 46-47	
15	Finish DNA sequencing analysis – Identify *Borrelia* strains	3f: 48	Tick-borne Pathogens Report Due Finals Week

Protocol Order:

Due at the beginning of each lab session, the following is required to be written in your lab manual, within the note section of each lab. This will consist of **headings & corresponding pages** from the lab manual in the order in which they will be conducted **for that day**. To minimize our waiting time, we will be conducting overlapping labs. One **group** will arbitrarily be chosen to write the protocol list on the board. The class will then decide whether or not it needs to be revised for that day.

Example
Week 1: Group #1
Prepare 1X TAE Buffer (p.3)
Prepare 1% Small Gel (p.3)

Laboratory Reports:

Genetic researchers generally perform multiple experiments on a daily basis. Therefore, it is very important that researchers keep excellent records of their experimental findings in laboratory notebooks. Lab books are a form of documentation of work that was performed and reported in published manuscripts. Your lab reports will be based on a revised version of a manuscript, whereas all of the detail of the introduction and methods sections will be omitted. However, the discussion and conclusion sections will be greatly emphasized. In order to keep accurate records during each lab investigation, it is critical that you document everything you do in a lab notebook. Although it will not be graded, it will help you to keep organized notes about each lab and collect the data for your reports.

Lab Reports are to be typed, 12 pt font, 1 inch margins all around, and written in the following format:

Title, Group #, and **Names** of your partners

Purpose: State the reason for doing the experiment (1-2 sentences)

Hypothesis: State your hypothesis that you are testing. Place in purpose section in formal lab reports.

Methods: In sentence form, include the general headings of each part of each investigation from your lab manual. You are basically citing your manual. Therefore, there is no need to write the detailed protocols over again. However, include any changes in the methods you may have performed.

Results: Summarize your data. Include labeled gel images, tables, graphs, DNA sequences, DNA fingerprints, genetic trees. Describe procedural problems that may have occurred. Include answering any questions from the results section of each investigation. Do not rewrite the questions.

Discussion: In paragraph form, answer the questions addressed in the discussion section of each investigation. Do not answer all of the questions using one long paragraph. Use separate paragraphs when addressing different topics. If you did not get PCR products, then discuss what could have gone wrong with your experiment and give suggestions on how to improve your technique and/or adjust the protocol, etc.

Conclusion: Interpret your results according to those expected and why unexpected results may have occurred. State how the exercise addressed the purpose stated above. State how your work relates to broader questions in genetics and any conclusions you can draw relating to your stated hypothesis. If you were to continue this project, what would be the next step? What questions need further explanation?