Syllabus and Schedule
BMB 303: Techniques in Biochemistry and Molecular Biology
Fall 2005-2006

Instructors: Dean Fraga, Biology, Mateer 304, x2557, dfraga@wooster.edu
Mark Snider, Chemistry, Severance 201, x2391, msnider@wooster.edu

Class: Mondays, 12-4 pm; Wednesdays 1-4 pm

Office Hours: By appointment

Texts:
1. "Protein Purification: Principles and Practice" by R. K. Scopes
2. "Applied Molecular Genetics" by R. L. Miesfeld
3. Handouts and web-based materials.

www.wooster.edu/biology/wmorgan/bmb303/

Laboratory Spaces

Class discussions will be held in Mateer 305 and in Mateer 308/310 for procedures involving molecular biology techniques. Severance 004 will be utilized for procedures involving protein biochemistry.

Description

This laboratory-based course will give students hands-on experience with some experimental methods used in biochemistry and molecular biology. It is organized around a semester-long project in which students will design and work towards specific research goals. This course counts for major credit in Biology and Chemistry. *Prerequisites:* Chemistry 112 and Biology 220. BMB majors are encouraged to have prior or concurrent enrollment in BMB 331.

Scope of the Course

The course will revolve around a central research project during which important laboratory techniques used in biochemistry and molecular genetics will be learned and performed. The research goal of this year's course is to explore the evolution of phosphagen kinases by studying these enzymes from bacterial sources. Using bioinformatics techniques, we will identify potential homologues from bacteria and clone the genes to study the biochemical properties of the resulting gene products.

General Outline

Students will work individually or in groups of two. The experiments will be performed in a logical order to address the research problem noted above. A tentative timetable is given below. However, since the course is project-oriented, a specific day-to-day timetable of experiments cannot be assigned at the outset. Students will be given several weeks to carry out each experiment. This will allow time for repeating experiments that fail due to "goofs" and periods set aside for experimental design, and discussion.

One faculty member will be the lead person during specific times corresponding to their expertise. Each instructor will lead the class at different times depending on the types of

experiments being performed. During these times, the other faculty member may occasionally visit labs and participate in discussions.

Grading

Grades will be assigned according to the following weighting scheme:

Quizzes:	40%
Lab reports (2):	40%
Final Proposal:	10%
Lab-book:	10%

Quizzes will be given weekly to test your knowledge of current experiments being performed. Quizzes will examine students' conceptual understanding of experimental design, methodology and ability to relate weekly results and ideas to the overall research goal. At the completion of both sections (molecular and biochemical) of the course, each student will prepare a formal laboratory report. Students will also keep a laboratory notebook that provides the details for each experiment performed. Guidelines for these assignments are described below and details will follow in handouts provided later in the semester. See the schedule below for deadlines. In place of a final exam each student will make an individual presentation to the class. Presentations will require students to explore the scientific literature and place the concepts learned in this course into a larger scientific perspective. More details will be provided later in the semester. These presentations will be given during the designated final exam time.

Tentative Schedule

Week	**Procedures Performed, Assignment Due Dates** ***Primary Instructor(s)***
1 8/30 & 9/1	Introduction and overview of research project. Use of Chimera and MacVector *Fraga/Snider*
2 9/6 & 9/8	Bioinformatics Plasmid structure and use. *Fraga*
3 9/13 & 9/15	Basics of recombinant DNA techniques Discussion of mutagenesis techniques. *Fraga*
4 9/20 & 9/22	DNA transformation technology Cloning technologies *Fraga*
5 9/27 & 9/29	Agarose gel electrophoresis. *Fraga*
6 10/4 & 10/6	DNA sequencing technology, genome projects, and bioinformatics. *Fraga*
7 10/11 & 10/13	Transformation of mutants into BL21 cell line. ***Lab Report Due 10/15*** *Fraga*
8 10/20	Protein expression and preparation of buffers. Protein purification: Harvesting bacteria *Snider*
9 10/25 & 10/27	Protein purification: Cell lysis and centrifugation Protein purification: Affinity Chromatography *Snider*
10 11/1 & 11/3	Protein purification:.Affinity Chromatography Protein purification: Affinity Chromatography *Snider*
11 11/8 & 11/10	Protein purification: SDS PAGE analysis Protein purification: SDS PAGE analysis *Snider*
12 11/15 & 11/17	Protein characterization: Enzyme kinetics, size exclusion chromatography, ligand binding assays *Snider*
13 11/22	Protein characterization: Enzyme kinetics, size exclusion chromatography, ligand binding assays *Snider*
14 11/29 & 12/1	Protein characterization: Enzyme kinetics, size exclusion chromatography, ligand binding assays *Snider*
15 12/6 & 12/8	Project conclusion and ideas for future directions ***Lab Report Due 12/6***. *Fraga/Snider*

Presentations (Final Exam Time): Friday 12/17 9:00 AM.

Laboratory Report Guidelines

Two formal laboratory reports will be completed during the course of the semester. These laboratory reports will be organized in a way that is standard to the presentation of scientific work. The sections and major features expected in each section of your laboratory report are given below. Use third person past tense in your writing. The use of "we" is acceptable, but should be used sparingly. Complete guidelines for the preparation of a lab report are detailed in Pechenik's *A Short Guide to Writing about Biology*. Published scientific papers, as well as what you have learned about science writing in Chemistry 211/212 also serve as good examples in how to write and structure your reports.

Title. Spend some time generating a title for your report. A good title should accurately describe the experiments carried out, giving as many specifics as possible in as few words as necessary.

Abstract. The abstract should explain concisely what the goal of the experiment was, what measurements were made, and a summary of the most important results obtained. The abstract must be one paragraph in length.

Introduction. In the Introduction, you should explain why you did the study and provide necessary background information to explain the significance of the work. Citation of research articles is required.

Materials and Methods. In this section, you should describe what you did in sufficient detail so as to permit your experiment to be repeated. However, the writing should be as concise as possible. For example, it is necessary to report the composition of the buffer you used, but it is not necessary to write that it was prepared using a 500 mL volumetric flask.

Results. In this section, your data are presented using tables, plots, and other graphics with accompanying text. It is important to point out any particularly interesting trends or data points. Please note that the Results section is:

1. <u>not the place</u> to discuss why the experiment was performed (covered in Introduction).
2. <u>not the place</u> to discuss how the experiment was performed (covered in Methods).

Finally, when appropriate, report the error associated with your data. This can typically be calculated using replicate measurements or other statistical means. Your instructor will point out when this type of analysis should be done.

Discussion. A thorough analysis of the data and conclusions derived from them should be made. The results must be interpreted in the context of the specific questions you set out to address as outlined in the introduction. Questions that should be answered in the Discussion include:

1. What was your hypothesis and what were the expectations of the designed experiments?
2. How did your results compare with those expected; do the results validate or invalidate your hypothesis?
3. What are possible, alternative explanations of the results and any unexpected results?
4. How might these alternative explanations be examined further by experimental means?

The Discussion is perhaps the most important section of your laboratory report. Length is not a factor in evalution, but depth of understanding and clarity in presentation is critical. Noting other techniques or methods that would be superior or may be helpful in addressing new questions is an example of good information to provide in the discussion.

Acknowledgments. If individuals other than those listed as authors helped in the investigation they should be specifically cited in the Acknowledgment section.

Literature Cited. Cite literature or manuals from which any information or procedures were derived when writing your report or performing the experiments described within.

Lab Notebook Guidelines

For advice on keeping an accurate and informative lab notebook, see "The purpose of laboratory and field notebooks" in Chapter 8, "Writing Laboratory and Other Research Reports," of Pechenik's *A Short Guide to Writing about Biology, 4th ed.*

For each experiment, your laboratory notebook should include the following as appropriate:

1. statement of objectives
2. experimental design
3. observations and data
4. analysis and interpretation
5. summary statement, including future objectives

Academic Integrity

All students are expected to read and honor the Code of Academic Integrity, which is printed in The Scot's Key. As stated in the preamble, adherence to the Code insures that the College is a community that promotes "the fullest learning by everyone."

The principles of the Code of Academic Integrity are excerpted here:
Under the Code of Academic Integrity, a student will not:
A. give, offer, or receive aid other than that specifically allowed by the professor on any course work or examination;
B. knowingly represent the work of others as his/her own; (This includes, but is not limited to, plagiarism...)
C. falsify data;
D. violate the spirit of the Code expressed in the Preamble.

Plagiarism is defined in the Code of Academic Integrity as follows:
To use or imitate the language, ideas, or thoughts of another person and represent them as one's own is to commit an act of plagiarism. This is true whether the material used is only a brief excerpt or an entire paper or article and whether the original source is the work of another student or some publication.

For a more complete description of plagiarism, consult *The Scot's Key* and *A Short Guide to Writing about Biology*. For examples of appropriate and inappropriate paraphrasing, consult *A Writer's Reference* (3rd ed., p. 261; on reserve in Gault/Andrews Library).

Note that while some assignments may be done in collaboration with a classmate, most should be done individually. *If you have any questions about your assignment, plagiarism, or the Code of Academic Integrity, always ask your instructor!*

Format to fit on 5 pages