

SYLLABUS

Biol.4290/5290 Recombinant Protein Production Techniques – Fall 2016

Lectures: T 12:30-2:10 PM; Location Olsen Hall 407
Labs: W 8:30 AM-12:20 PM; Location Olsen Hall 509
or R 12:30 PM-4:20 PM; Location Olsen Hall 509

Instructors:

Prof. Hwai-Chen Guo; Olsen 413B, ext. 4-2878, HwaiChen_Guo@uml.edu
TA: Suchita Pande; Olsen 508, ext. 4-5072, Suchita_Pande@student.uml.edu

Course Prerequisites:

Experimental Methods in Biology (Biol.2330L) or equivalent

Course Materials:

Required Text*: Protein Purification: Principles, High Resolution Methods, and Applications. J-C Janson (2011), 3rd Ed. (hardcopy: ISBN 978-0-471-74661-4; e-book: ISBN 978-1-118-00219-3), John Wiley & Sons, Inc.; **2nd Ed. is ok to save money.**

Supplemental slides from sources outside of the textbook are provided in this booklet.

Lab procedures are distributed in a separate handout. These procedures should be read by each student prior to coming to each laboratory session.

Calculator – You will need a calculator that is capable of doing simple calculations.

Other: A bound laboratory notebook need to be purchased by the students.

Course Description:

This course introduces students to the principles and practice of recombinant protein expression and purifications. Proteins are major targets of pharmaceuticals, and are themselves increasingly used as therapeutics. However both basic research and pharmaceutical industry depends on availability of purified proteins that are often difficult to isolate from native sources. This course will provide both didactic and laboratory instruction. It is comprised of a series of lecture and laboratory exercises, with an emphasis on practical techniques and hands-on experience of recombinant protein purification. The course will cover a variety of expression systems, including prokaryotic and eukaryotic cells, and address traditional and new methods in protein purification.

Learning Objectives:

1. Develop a basic knowledge about recombinant protein production.
2. Demonstrate an understanding of gene manipulation to enhance protein expression.
3. Obtain a working knowledge of various expression systems.
4. Identify major approaches in protein purification.
5. Gain a hands-on experience of protein purification techniques.

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Grading Policies:

Students will be graded on the evaluation of two 75-min exams, the final take-home exam/report, weekly lab reports, and a combination of class participation, attendance, and overall attitude toward the course.

Examinations

Students will be examined through two mechanisms:

1. Two 75-min essay style written examinations designed to test the students' knowledge of individual aspects of protein expression and purification. The exams will have open-response questions, and may require students to utilize the information from class to design experiments to address problems, or to interpret hypothetical experimental data. These examinations will constitute thirty percent (30%) of the final grade.
2. A written term paper (take-home final exam) based on a detailed publication in the recent literature. The publication will be chosen by the student in consultation with the course instructor and contain significant discussion and use of protein expression/purification. Students need to carefully choose their papers, and are advised to seek the input of the instructor by mid-semester as to the appropriateness of a chosen paper. It must be a primary research article or letter from a peer-reviewed scientific journal, such as *Nature Methods*, *J. Biol. Chem.*, *Protein Expression and Purification*, or *J. Chromatog.*, and is published within the past 18 months, or research that you have performed or are currently performing at UML (or elsewhere). In the written term paper the student will be expected to include a research review on that topic, analyze, present the required theoretical and experimental approach, and criticize the results obtained within the context of limitations in the methods. The report will be evaluated based on the relevance and difficulty of the chosen paper as well as the thoroughness of the discussion. This report will constitute twenty percent (20%) of the final grade.

For students taking the graduate credit (Biol.5890)

Students will be evaluated as for undergraduate students, except with an additional requirement of presenting their term paper, typically by Microsoft Powerpoint presentation or its equivalent. All graduate students will make a 12 minute oral presentation at the end of the course, in a format similar to those presented at professional meetings, and critique each other's presentations. Students need to distribute copies of their papers to the class at least two weeks before their presentations. Presentations will be evaluated by the instructor and your peers (using a scoring sheet to be distributed) based upon the relevance and difficulty of the chosen paper as well as the thoroughness of the presentation. Students are expected to provide enough background information so that the audience would understand why and how the research was done, and what is the scientific gap or hypothesis the paper was intended to address. Students are also expected to provide their own critiques/opinions of the paper.

Grading Weights:

	<u>Undergraduates</u>	<u>Graduates</u>
Exam I	15%	15%
Exam II	15%	15%
Final Exam/Report	20%	20% (incl. oral presentation)
Weekly Pre/Post-Lab Reports	40%	40%
Participation	10%	10%

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Course Rules:

The Course Syllabus and the Course Rules are subject to change throughout the semester at the discretion of the instructor. Timely notice will be given regarding any changes, either in class, by e-mail, and/or on the website
http://faculty.uml.edu/hwaichen_guo/Teaching/Teaching.aspx

As in any classroom or research laboratory setting, standards of common courtesy and behavior are expected. Included among these are the following:

In Class/Lab:

- Be punctual and turn off mobile communication devices
- No cell phones, iPod, IM, nor chatting
- No food or beverages are allowed
- Plan, in advance, to remain in the lecture/lab for the duration of the class time
- Be respectful of the instructor and other students, ask questions but no chatting
- Attire during lab - closed-toes shoes are mandatory; clothing must fully cover your arms and legs; leave equipment clean and in good condition; return items to their appropriate location so they can easily be found by the next people who need to use them.

In Class/Presentation:

- No Cell phones, iPod, IM
- Graduate students have 12 minutes for their presentations
- Undergraduates need to attend the graduate presentations and participate in the peer evaluation
- Missed presentations cannot be make-up
- Be respectful of other students' presentations, ask questions but no chatting

Exams:

- Closed book for exams I & II; Open book/take-home for the final exam/report
- No make-up exams

Attendance:

Because this is a practical laboratory course, attendance and execution of the experiments are compulsory. Labs cannot be made up (except due to weather-related cancellation). If you miss a lab and have appropriate documentation indicating a valid reason for missing lab, you will be excused from that lab. If you do not have documentation indicating a valid reason for missing lab, you will be given a zero for that post-lab report. You cannot use your partner's or anyone else's data to submit a post-lab report for a lab you missed.

Format of Post-lab Reports:

The **post-lab report** for each lab must be turned in **on the following week right before the next laboratory starts**. There will be a receptacle box on a table, just inside room 509 for the post-labs at the beginning of each lab. The box will be removed 10 minutes following the start of the lab and the post-labs removed for grading. Turning in the lab reports after this time will not be permitted. One formal post-lab report will be prepared using the laboratory data in your lab notebook, and should cover lab objectives, importance, methods, calculations, results and discussions. Quality of work will be assessed based on results obtained, a proper recording of raw data onto the lab notebook, and the thoroughness of the discussion.

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Academic Honesty:

Cheating or plagiarism on any exam or report will not be tolerated, and could receive an “F” for the course

Any student caught cheating or helping another student cheat will earn zero for that assignment. For more information see:

<https://www.uml.edu/Catalog/Undergraduate/Policies/Academic-Policies/Academic-Integrity.aspx>

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Lecture Schedule:

Dates	Subject	Chapter*
September 6 T	1: Course Intro, Recombinant DNA Technology	handouts
13 T	2: Manipulation of Gene Expression in Prokaryotes	handouts
20 T	3: Intro to Protein Purification and Chromatography	1,2
27 T	4: Affinity	9
October 4 T	5: Ion Exchange	4
11 T	No Class, Monday Class Schedule	
18 T	Exam I (lectures 9/6 – 10/4)	
25 T	6: Heterologous Protein Production in Eukaryotes	handouts
November 1 T	7: Immobilized Metal Ion; 8: Gel Filtration	7,3
8 T	9: Reversed-Phase and Hydrophobic Interaction	5,6
15 T	10: Refolding of Inclusion Body	13
22 T	Exam II (lectures 10/25 – 11/15)	
29 T	Graduate Student Presentations	papers
December 6 T	Graduate Student Presentations	papers
12 M	Final Exam due (take-home exam/report)	

* chapter in the required textbook

Lab Schedule:

Dates	Subject	Lab
September 7/8	organization; pipetting exercises; agar plate preparation	1
14/15	bacterial transformation; stock solutions	2
21/22	inoculation, induction of expression, and cell harvest	3
28/29	isolation, separation, and prep-scale affinity column	4
October 5/6	enzyme activity assay; protein quantification	5
12/13	No Lab, Columbus Day Week	
19/20	crystallization test of purified proteins	6
26/27	insect cell techniques, and baculovirus-mediated expression	7
November 2/3	cell harvest, separation, and His-tag affinity column	8
9/10	FPLC gel filtration (groups 1-4); prepare SDS gel	9
16/17	FPLC gel filtration (groups 5-8); prepare SDS gel	9
23/24	No Lab, Happy Thanksgiving	
Nov 30/Dec 1	PAGE analysis	10
December 7/8	lab make-up if necessary	