

METHODS: ANALYSES**BIO 6120****SYLLABUS, WINTER, 2017**

Credits: 4

Time and Place: W/F 11:20-2:20 Biological Sciences, Room 4156

Instructor: Dr. Penelope I. Higgs
Biological Sciences Building, Room 4121
Phone: 313 577 9241

Goals: Introduction to essential principles in design, execution and analysis of experiments with proteins. Topics that will be covered includes laboratory safety, scientific documentation, development of experimental protocols, error analysis, solutions and buffers, over-expression and purification of proteins, electrophoretic separation of proteins, immunoblots, isoelectric focusing, protein-protein interactions, electrophoretic mobility shift assays and scientific ethics.

Limited to 20 students.

Prereq: BIO 5330 or BIO 6330 or consent of instructor.

Material fee: \$50.00

Format: Lectures and lab practicals accessible at:

Lab safety requirement: Lab coat

Contact and communication after lecture or via email: pihiggs@wayne.edu

Office hours: After lecture or by appointment

Textbook: handouts

Supplementary Textbooks (not required):

Kathy Barker: At the Bench: A Laboratory Navigator (Spiral-bound)

Safety Sense: A Laboratory Guide, Second Edition by Cold Spring Harbor Lab

TOPICS COVERED

1. Laboratory safety rules
2. Lab notebook management
3. Recombinant protein overexpression
4. Protein affinity purification
5. Immunoblot analysis
6. Co-purification of interacting proteins
7. Bacterial/Yeast Two-hybrid analyses
8. Isoelectric focusing
9. Electrophoretic mobility shift analyses
10. Purification of interaction partners from lysates

LEARNING OBJECTIVES/OUTCOMES

As a result of mastering the materials in this course, you will be able to:

1. Conduct laboratory research under supervision
2. Generate proper documentation of experimental results for academic and industrial settings keeping notebook
3. Perform basic protein purification experiments
4. Perform basic protein characterization experiments
5. Analyze protein architecture in silico
6. Discuss and analyze experimental results

Exams: Class performance will in part be measured in form of one quiz per week. The lowest scoring quiz (this includes missed quizzes) can be dropped. There will be **NO** makeup exams or bonus points.

Grading: 40% of the final grade will be based on the average score determined for the weekly quiz results. 50% of the final grade will be based on accuracy and organization of the laboratory notebook in which you will document your experimental work during the lab sessions. The laboratory notebook will be submitted for grading after each lab class and returned during lecture each following Monday. 10% of the final grade will be based on presentations and participation in the class.

Cheating policy: A student found to be cheating during an exam (using a “cheat sheet”, looking at another’s paper, or allowing another to look at yours) will receive a zero for that test or report with no opportunity to drop or replace that score. A second episode of cheating will result in a grade of F for the course and may also result in initiation of university disciplinary action.

Add/Drop policy: Add forms will not be signed after the second week of class (except for the purpose of changing lab sections when and if appropriate). Drop forms must be signed before the end of “study day”, which is the day after the last day of classes. Note that **“incomplete”** grades will not be issued to students in poor standing who are seeking an alternative to late drop.

Students with disabilities: If you have a documented disability that requires accommodations, you will need to register with Student Disability Services for coordination of your academic accommodations. The Student Disability Services (SDS) office is located at 1600 David Adamany Undergraduate Library in the Student Academic Success Services department. SDS telephone number is 313-577-1851 or 313-577-3365 (TDD only). Once you have your accommodations in place, I will be glad to meet with you privately during my office hours to discuss your special needs. Student Disability Services' mission is to assist the university in creating an accessible community where students with disabilities have an equal opportunity to fully participate in their educational experience at Wayne State University.

Credit requirement policy: Note that prerequisite requirements will be strictly enforced except for cases of extreme urgency, which will be decided on at the instructor's discretion.

class	date	day	topic	student activities	instructors	homework
1	1/11/17	Wed	introduction- isolate and transform expression plasmids	isolate plasmids	transform to induction strains	
2	1/13/17	Fri	Recombinant protein expression	induce cells, prepare samples		
3	1/18/17	Wed	Recombinant protein expression cont.	prepare gels / run gels / (stain gels)	(stain gels), post to blackboard	
4	1/20/17	Fri	Recombinant protein solubility	prepare samples / run gels / (stain gels)	prepare gels / (stain gels), post to blackboard	lab notebook
5	1/25/17	Wed	Western Blot	prepare gels/run gels/ transfer	membrane in blocking buffer	
6	1/27/17	Fri	Western Blot cont.	perform WB / (expose to film)	(expose to film), post to blackboard	in class quiz
7	2/1/17	Wed	Recombinant protein purification	induce large cultures, pellet	prepare o/n cultures	
8	2/3/17	Fri	Recombinant protein purification cont.	affinity purify recombinant proteins	lyse cells, prepare columns	bioinformatics exercise; lab notebc
9	2/3/17	Wed	Recombinant protein purification cont.	Run gels analyze results	dialyze / freeze samples /	
10	2/8/17	Fri	Native gel electrophoresis	prepare gels		quiz
11	2/10/17	Wed	Native gel electrophoresis	run gels	post to blackboard	
12	2/15/17	Fri	IEF	lecture		lab notebook
13	2/17/17	Wed	IEF	1-D		
14	2/22/17	Fri	IEF	2nd-D, blot		quiz
15	2/24/17	Wed	IEF	Western blot		
16	3/1/17	Fri	EMSA	lecture		quiz; lab notebook
17	3/3/17	Wed	EMSA	prepare gels		
18	3/3/17	Fri	EMSA	run gels		quiz
19	3/8/17	Wed	EMSA	analyze data		
20	3/10/17	Fri	EMSA / copurification	student presentations / lecture		quiz; lab notebook
	3/15/17	Wed	spring break			
	3/17/17	Fri	spring break			
21	3/22/17	Wed	Protein co-purification	Lecture & prepare gels		experimental design for protein pu
22	3/24/17	Fri	Protein co-purification	prepare samples / run gels / (stain gels)		quiz; lab notebook
23	3/29/17	Wed	Protein co-purification	Western blot		
24	3/31/17	Fri	B2H/V2H	transform		quiz; lab notebook
25	4/5/17	Wed	B2H/V2H	plate		
26	4/7/17	Fri	B2H/V2H	data analysis		quiz; lab notebook
27	4/12/17	Wed	protein purification project			
28	4/14/17	Fri	protein purification project			
29	4/19/17	Wed	protein purification project			
30	4/21/17	Fri	protein purification project			