

CHM 4300L – Laboratory in Biochemistry and Molecular Biology

Instructor

Alix Rexford

Zoom ID: 277-621-8481

alix.rexford@chem.ufl.edu

Office hours via Zoom: Tuesday 2:00 – 4:00 PM and by appointment

Laboratory Manual

Characterization of TEM1 β -Lactamase and Discovery of Inhibitors from Streptomyces

Available at Target Copy Center

Class Meetings

Pre-laboratory synchronous lecture: Wednesday 10:40 – 11:30 AM; Access Zoom through Canvas
Laboratory: 11392: Thursday 9:35 – 12:35 PM; Leigh Hall 200/300
11393: Thursday 12:50 – 3:50 PM; Leigh Hall 200/300

Course Description

This course provides a practical, hands-on understanding of modern, fundamental techniques relevant to molecular biology and biochemistry. The laboratory covers topics including DNA cloning and manipulation, basic bioinformatic analyses, protein overexpression and purification, along with enzyme kinetic measurements. Additionally, this course covers the discovery of enzyme inhibitors and antibiotics from natural sources.

Laboratory Attire & COVID-19 Safety Precautions

Students should wear goggles, gloves, and closed toe shoes with hair pulled back at all times while in the laboratory. No shorts, loose clothing, or jewelry are allowed. In class laboratory sessions are reserved for experiments only; data analysis, lab write ups, and discussions will be done outside the lab. In response to COVID-19, the following policies and requirements are in place to maintain your learning environment and to enhance the safety of our in-classroom interactions.

- You are required to wear approved face coverings at all times during class and within buildings. Following and enforcing these policies and requirements are all of our responsibility. Failure to do so will lead to a report to the Office of Student Conduct and Conflict Resolution.
- This course has been assigned laboratory rooms with enough capacity to maintain physical distancing (6 feet between individuals) requirements. Please utilize designated stations and maintain appropriate spacing between students. Do not move from the station you are assigned.
- Sanitizing supplies are available at each station to wipe down your lab bench at the start and end of the lab. Hand soap will be provided at each sink and should be used prior to lab beginning and before exiting the laboratory.
- Practice physical distancing to the extent possible when entering and exiting the classroom and when using common-use equipment. Sanitizing solutions will be available to clean common-use equipment before and after each use.
- If you are experiencing COVID-19 symptoms ([Click here for guidance from the CDC on symptoms of coronavirus](#)), please use the UF Health screening system and follow the instructions on whether you are able to attend class. [Click here for UF Health guidance on what to do if you have been exposed to or are experiencing Covid-19 symptoms](#).
 - Course materials will be provided to you with an excused absence, and you will be given a reasonable amount of time to make up work. [Find more information in the university attendance policies](#).

Honor Code

I expect each of you to follow the Student Honor Code, available on the web (<https://sccr.dso.ufl.edu/policies/student-honor-code-student-conduct-code/>)

You are expected to:

- a. uphold the highest standards of academic integrity in the student's own work,
- b. refuse to tolerate violations of academic integrity in the University community
- c. foster a high sense of integrity and social responsibility on the part of the University community.

Violations of the Honor Code will be reported to the Dean of Students, and may result in failure of the assignment in question and/or the course.

Laboratory Attendance

Attendance is required for all lab sessions. **Please be on time!** Your performance grade depends on coming to lab on time, **with proper safety attire**, and having read the experiment thoroughly in advanced. There will be 2 – 3 short recorded lectures followed by questions to check your understanding. This **MUST** be completed prior to attending lab for your own safety, as well as to ensure that everyone understands proper procedure. Proper preparation will allow you to work quickly to complete the lab in a timely manner. Due to the continuity of the labs in the course, missed labs cannot be made up. If you miss a lab due to **an approved absence with appropriate documentation**, accommodations will be made.

Assignments

E-Notebooks will be graded for accuracy and completeness. Notebooks should include a short description of the experiments including the scientific concepts of the experiment, all data collected during the laboratory period, and any data analysis. To reduce the amount of things brought into lab, lab notebooks will be done electronically. You may bring a laptop or tablet into the laboratory to be kept in a designated area on the lab benches. **Nothing else should be brought into the laboratory.**

To make the most of our class time, you will have 15-30 minutes of pre-lab **videos to watch prior to Wednesday's synchronous lecture**, followed by a short **quiz to check your understanding**. During the **synchronous lecture** we will review concepts from the previous week and discuss issues and results, as well as potential problems that may arise in the following week. After a short class discussion, students will work together on editing their manuscripts.

Throughout the semester you will be preparing a **journal-quality manuscript** encompassing the semester's project. Every other week you will prepare one section of the paper and submit it no later than 11:59 pm on Monday. Documents will be shared with your lab partners via Canvas, and in class on Wednesday you will discuss the section of the manuscript.

Paper section	Group	Rough Draft Due	Final Draft Due
Introduction	A	Sept. 7	Sept. 18
Introduction	B	Sept. 14	Sept. 18
Methods (labs 1-2 + virtual labs 1-2)	A	Sept. 21	Oct. 2
Methods (labs 1-2 + virtual labs 1-3)	B	Sept. 28	Oct. 2
Results (labs 1-3 + virtual labs 1-3)	A	Oct. 5	Oct. 16
Results (labs 1-3 + virtual labs 1-4)	B	Oct. 12	Oct. 16
Discussion (labs 1-4 + virtual labs 1-4)	A	Oct. 19	Oct. 30
Discussion (labs 1-4 + virtual labs 1-5)	B	Oct. 26	Oct. 30
Abstract	A	Nov. 2	Nov. 20
Abstract	B	Nov. 16	Nov. 20

After class, you may edit your paper section according to your class discussions and turn in a final draft by the due date listed above. These will be graded according to the provided rubric. You are expected to make the recommended alterations to each section as well as include all laboratories for your final manuscript, which is due December 7, 2020.

Grading

Laboratory notebooks	200 pts	
Quizzes	100 pts	
Lab performance	40 pts	
Synchronous lecture participation	40 pts	
Lab report sections	120 pts (20 pts each)	
Final lab report (labs 1-6 + virtual labs 1-6)	100 pts	Due Dec. 7

The following letter grades will be assigned based upon total points accrued for all course work:

A: 600 – 540 pts

B: 539.9 – 480 pts

C: 479.9 – 420 pts

D: 419.9 – 360 pts

F: below 360 pts

Students with Disabilities

Students requesting classroom accommodation must first register with the Dean of Students Office. The Dean of Students Office will provide documentation to the student who must then provide this documentation to the Instructor when requesting accommodation. Contact the Disability Resources Center (<http://www.dso.ufl.edu/drc/>) for information about available resources for students with disabilities.

Course Evaluations

Students are expected to provide professional and respectful feedback on the quality of instruction in this course by completing course evaluations online via GatorEvals. Guidance on how to give feedback in a professional and respectful manner is available at gatorevals.aa.ufl.edu/students/. Students will be notified when the evaluation period opens and can complete evaluations through the email they receive from GatorEvals, in their Canvas course menu under GatorEvals, or via ufl.bluera.com/ufl/. Summaries of course evaluation results are available to students at gatorevals.aa.ufl.edu/public-results/

Date	Lab	Group	DNA/Protein Biochemistry	<i>Streptomyces</i> Microbiology	Virtual Lab	Description
Sept 3					V1 – Group A & B	Designing PCR primers
Sept 10	1	A	DNA agarose gel, DNA cleanup & restriction digest	Plate soil samples	V2 – Group B	DNA Bioinformatics
Sept 17	2	B	DNA agarose gel & cleanup, DNA quantitation	Select <i>Streptomyces</i>	V2 – Group A	DNA Bioinformatics
Sept 24	3A	A	Ligate DNA & transform into TOP10 cells	Re-streak <i>Streptomyces</i>	V3 – Group B	Recombinant plasmid digest and gel
Oct 1	3B	B	Ligate DNA & transform into TOP10 cells	Prepare <i>Streptomyces</i> plate for antibacterial test	V3 – Group A	Recombinant plasmid digest and gel
Oct 8	4A	A	TEM-1 purification from BL21(dE3) cells	Spot bacterial test strains for antibacterial test	V4 – Group B	Expression testing and SDS-PAGE
Oct 15	4B	B	TEM-1 purification from BL21(dE3) cells	Start <i>Streptomyces</i> liquid culture/Frozen stock	V4 – Group A	Expression testing and SDS-PAGE
Oct 22	5A	A	Purification tests	Isolate/wash liquid culture resin	V5 – Group B	Visualizing protein structure (Pymol)
Oct 29	5B	B	Purification tests	Extract metabolites w/methanol	V5 – Group A	Visualizing protein structure (Pymol)
Nov 5	6A	A	Inhibition assays	Kinetics w/TEM-1 & Ab test	V6 – Group B	Molecular docking
Nov 12	NO CLASS					
Nov 19	6B	B	Inhibition assays	Kinetics w/TEM-1 & Ab test	V6 – Group A	Molecular docking