

CHM 4300L – Laboratory in Biochemistry and Molecular Biology

Instructor

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Office hours: Monday 3:00 – 4:00 PM; Wednesday 1:00 – 2:00 PM; Or by appointment

Laboratory Manual

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

Class Meetings

Pre-laboratory synchronous lecture: Tuesday 11:00 – 12:15 PM; Access Zoom through Canvas
Laboratory: Monday 9:30 – 12:15 PM; CCB 110

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, protein overexpression and purification, along with enzyme kinetic measurements as well as the discovery of enzyme inhibitors and antibiotics from natural sources.

Additionally, this course provides instruction and feedback to help students improve their scientific writing. At the conclusion of this course students will have composed a journal-article quality manuscript regarding their experimental findings in the laboratory.

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. Failure to follow safety precautions and proper use of PPE will result in your removal from the laboratory.

- 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.
- All students in face-to-face courses are required to test for COVID every 2 weeks in order to maintain campus clearance. If you are not cleared to attend campus, you will be removed from the laboratory. A virtual laboratory will be assigned in lieu of the wet laboratory.
- 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 Participation

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. All students will begin the semester with 50 laboratory participation points. Five (5) points will be deducted for each laboratory period in which a student fails to attend without a valid excuse presented to the instructor no later than the start time of that laboratory period. Four (4) points will be deducted any time a student fails to clean up and sign the log sheet at the end of each laboratory. Three (3) points will be deducted for each tardy of ten minutes or more. Points may also be deducted for gross negligence in the lab or major errors in following protocols, at the discretion of the instructor.

There will be 2 – 3 short recorded lectures followed by questions to check your understanding which **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.

Lecture Participation

Attendance is required for all synchronous Zoom lecture meetings. 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 and/or data analysis. Your active participation in these small group sessions will determine your lecture participation score.

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 and all reaction components and conditions, all data collected during the laboratory period, and any data analysis as indicated in the lab manual. To reduce the amount of things brought into lab, lab notebooks will be completed electronically via OneNote. 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.**

Select **data analyses** will be turned in via Canvas and graded on a pass/fail basis. You will have at least one (1) chance to correct your data analyses before grades are final, however, you **MUST** turn in completed data analyses on the initial due date. Failure to attempt **ALL** parts of the assignment will result in a score of zero

(0).

To make the most of our class time, you will have 15-30 minutes of pre-lab **videos to watch prior to lab**, 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. You will prepare one section of the paper and submit it to your OneNote collaboration space by the due date indicated in the table below. You will discuss and edit your classmates' drafts of the manuscript during specified lecture days.

<i>Manuscript Section</i>	Labs to be included	Rough Draft Due Date	Final Draft Due Date
<i>Introduction</i>	N/A	May 17	May 25
<i>Methods</i>	Labs 1-4	May 31	June 15
<i>Results</i>	Labs 1-6	June 28	July 6
<i>Discussion</i>	Labs 1-8	July 12	July 27
<i>Full Manuscript</i>	Labs 1-9	---	August 2

*Labs listed in the table above are **REQUIRED** to be included in your *final draft of the specified section*, although the most recent lab may be excluded in the rough draft as you won't have completed it prior to the rough draft due date. I would **HIGHLY recommend including Lab 9 in your discussion final draft**, as the bulk of the discussion comes from this lab and you will receive better feedback prior to your submitting your full manuscript.

After class, you should 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 and an abstract for your final manuscript, which is due August 2, 2021. Failure to make recommended changes for the final manuscript will result in a score no more than five (5) points less than received on the section final draft (not including rough draft points).

Tentative lecture schedule:

<i>Date</i>	<i>Discussion Topics</i>	<i>Assignment</i>	<i>Due Date</i>
May 11	Project overview Introduction Section PowerPoint	Lab overview Quiz	May 11 @ 10:55 am
May 18	Introduction rough draft – peer edits	PCR product data analysis	May 20 @ 11:59 pm
May 25	Method Section PowerPoint	PCR primers Problem Set	June 7 @ 11:59 pm
June 1	Methods rough draft – peer edits Complete Problem Sets	Restriction digestion Problem Set	June 7 @ 11:59 pm
June 8	Experimental overview Methods rough draft – peer edits		
June 15	Results Section PowerPoint	Expression conditions Problem Set	June 28 @ 11:59 pm
June 21	No Class – Summer Break		
June 29	Results rough draft – peer edits		
July 6	Discussion Section PowerPoint Results rough draft – peer edits		
July 13	Discussion rough draft – peer edits		

July 20	Discussion rough draft – peer edits	Purification table	July 20 @ 11:59 pm
July 27	Abstract Section PowerPoint	Kinetic data analysis	July 27 @ 11:59 pm

Late Assignments

Laboratory notebooks will be kept on OneNote. Grading dates will be announced, but since it is not “turned in” it cannot be turned in late. Please keep up with your notebook weekly.

Quizzes must be completed prior to the start of lab for your safety. There are no makeups for pre-lab quizzes.

Report section rough drafts are not graded, but **turning it in on time** counts towards the section’s final draft score. Rough drafts must be written in full sentences and follow the guidelines set forth in the powerpoint descriptions and lab report grading rubric to earn credit in the final draft score.

Late assignments for report section final drafts and the final manuscript are accepted with the following grade penalties:

10 % deduction: < 24 hours after due date

20 % deduction: 24 – 48 hours after due date

50 % deduction: 48 – 72 hours after due date

Not accepted after 72 hours late

*You almost never make up the points lost from turning it in late. I don’t recommend doing it!

Grading

Laboratory notebooks	200 pts	
Quizzes	100 pts	
Lab performance	50 pts	
Synchronous lecture participation	50 pts	
Lab report sections	100 pts (25 pts each)	
Full Manuscript (labs 1-9)	100 pts	Due Apr. 20

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

Regrades and Grading Errors

Mistakes happen and grading errors can be especially frustrating. I will do my best to make sure that you are always kept up to date with your performance in the course and post your grades in a timely manner. It is your responsibility to make sure that your grade on Canvas reflects the scores you receive on assignments and bring it to my attention ASAP when you notice a discrepancy. If you feel that a grading error was made on a particular assignment you have **one week from the date the grade was posted to Canvas** to request a regrade. Regrades will include a rescore of the entire assignment and may result in a lower score.

Student Responsibilities & Zoom Etiquette

You are expected to come to class on time and behave in a manner that is respectful to the instructor and to

fellow students. Opinions held by other students should be respected in discussion, and conversations that do not contribute to the discussion should be held at a minimum.

To add a profile picture go to: ufl.zoom.us/profile

To ask a question in Zoom: Click on the “Chat” button on the bottom of the screen and a chat window will appear

To raise your hand in Zoom: Click on the “Participants” button on the bottom of the screen and a window will appear. On the lower right of the window there is an option to raise your hand.

Privacy Statement

Our class sessions will be audio visually recorded for students in the class to refer back. Students who participate with their camera engaged or utilize a profile image are agreeing to have their video or image recorded. If you are unwilling to consent to have your profile or video image recorded, be sure to keep your camera off and do not use a profile image. Likewise, students who un-mute during class and participate orally are agreeing to have their voices recorded. If you are not willing to consent to have your voice recorded during class, you will need to keep your mute button activated and communicate exclusively using the "chat" feature, which allows students to type questions and comments live. The chat will not be recorded or shared. As in all courses, unauthorized recording and unauthorized sharing of recorded materials is prohibited. Recordings will not normally be made during breakout sessions, however those that are made are for the sole use of the instructor and will not be made available to students or others.

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/

U Matter, We Care

Your well-being is important to the University of Florida. The U Matter, We Care initiative is committed to creating a culture of care on our online campus by encouraging members of our community to look out for one another and to reach out for help if a member of our community is in need. If you or a friend is in distress, please contact umatter@ufl.edu so that the U Matter, We Care Team can reach out to the student in distress. A nighttime and weekend crisis counselor is available by phone at 352-392-1575. The U Matter, We Care Team can help connect students to the many other helping resources available including, but not limited to, Victim Advocates, Housing staff, and the Counseling and Wellness Center. Please remember that asking for help is a sign of strength. In case of emergency, call 911.

Tentative Laboratory Schedule (Dates subject to change)

<i>Date</i>	<i>Lab</i>	<i>DNA/Protein Biochemistry</i>	<i>Streptomyces Microbiology</i>
May 10	1	Introduction to pipets; Polymerase chain reaction	Plate soil samples
May 17	2	DNA agarose gel, PCR cleanup & restriction digest	Select <i>Streptomyces</i>
May 24	3	DNA agarose gel & cleanup, DNA quantitation; Ligate DNA	Re-streak <i>Streptomyces</i>
May 31	NO LAB	Memorial Day Holiday	
June 7	4	Transform TOP10 cells with Ligation Reactions; Miniprep Plasmid DNA	Prepare <i>Streptomyces</i> plate for Antibacterial Test
June 14	5	Restriction Digest Analysis; Transform BL21(dE3) Cells with Recombinant Plasmid	Spot bacterial test strains for Antibacterial Test
June 21	NO LAB	Summer Break Holiday	
June 28	6	Pilot-Scale TEM1 Expression Trials	Start <i>Streptomyces</i> Liquid Culture; Create Frozen stock
July 5	NO LAB	Independence Day Holiday	
July 12	7	TEM-1 Purification from BL21(dE3) cells	Isolate/wash Resin from Liquid Culture
July 19	8	Purification tests	Extract Metabolites from Resin
July 26	9	Inhibition assays	Kinetics w/TEM-1 & Antibacterial test