

CHM 4300L – Laboratory in Biochemistry and Molecular Biology

Instructor

Alix Rexford

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Learning Assistants:

Kanishka Ekanayake

Jorge Dominguez

Salma Drew

Colleen Martin

Nayda Paz Duany

Dr. Rexford's Office hours:

Monday 1:00 – 2:00 PM

Wednesday 12:30 – 1:30 PM

Or by appointment; Office hours are held via HyFlex; see syllabus for Zoom link

LA office hours (Zoom only)

Laboratory Manual (available at Target Copy)

Characterization of TEM1 β -Lactamase and Discovery of Antibacterial Metabolites from Streptomyces

Class Meetings

Pre-laboratory lecture: Wednesday 10:40 – 11:30 AM TUR2333

Laboratory: Meet in LEI 200

Section	Day & time	Teaching Assistant
BLR3 (ThurAM)	Thursday @ 9:35 - 12:35 PM	Nessa Pesaran Afsharian
B3R6 (ThurPM)	Thursday @ 12:50 - 3:50 PM	Peter Yang

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, and 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-quality manuscript regarding their experimental findings in the laboratory.

Laboratory Attire & COVID-19 Safety Precautions

Students should wear safety glasses, gloves, and closed toe shoes with hair pulled back at all times while in the laboratory. Shorts, loose clothing, or jewelry are prohibited. 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.

If you are experiencing COVID-19 symptoms ([Click here for guidance from the CDC on symptoms of coronavirus \(Links to an external site.\)](#)), 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 \(Links to an external site.\)](#).

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 any time a student fails to clean up their station at the end of each laboratory. Five (5) 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 – 4 short, pre-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. Missing a lab without an approved absence will result in a 10 point deduction from your course grade for the first absence, 40 point deduction for the second absence, and 100 point deduction for the third absence. Regardless of whether or not the absence is excused, you are still responsible for completing pre-lab quizzes and lab notebooks (using lab partner's data) by applicable deadlines.

Lecture Participation

Attendance is required for all lecture meetings on Wednesdays, 10:40 – 11:30 am. During the **lecture period** we will review concepts from the previous week and discuss theoretical concepts 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, complete problem sets, and/or data analysis. Your active participation in these small group sessions will determine your lecture participation score.

All students will begin the semester with 50 lecture participation points. Three (3) points will be deducted for each tardy of five minutes or more. Five (5) points will be deducted for each unexcused absence after the first unexcused absence. Points may also be deducted for lack of participation and discussion with your group, at the discretion of the instructor.

Assignments

OneNote will be utilized for E- lab notebooks, which will be graded for accuracy and completeness. Set up your notebook by navigating to the Class Notebook tab and opening the OneNote notebook. Each lab will have a dedicated page with an included grading rubric. You **MUST** use these pages to submit all lab notebooks. The lab notebook grading rubric provides an overview of how to complete each page and upload a document to OneNote, if desired. Do not copy and paste any text or figures into OneNote.

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 **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 **publication-quality manuscript** encompassing the semester's project. You will prepare a rough draft of each section of the paper and submit it to Canvas by the due date indicated in the table below.

Manuscript Section	Labs to be included	Rough Draft Due Date	Final Draft Due Date
Introduction	N/A	January 13	January 24
Methods	Labs 1-4	January 27	February 14
Results	Labs 1-8	February 17	March 7
Discussion	Labs 1-10*	March 24	April 4
Full Manuscript	Labs 1-11	---	April 24

*Full data analysis of Lab 10 will not be complete by the deadline. Discussion must include Lab 10 Part 2 analysis, but may omit Lab 10 Part 1 analysis

Labs listed in the table above are **REQUIRED** to be included in your **final draft of the specified section**, although the most recent lab(s) may be excluded in the rough draft as you won't have completed it prior to the rough draft due date.

After submitting your rough draft on Canvas, you will be randomly assigned two (2) anonymous **peer reviews**. Instructors do not grade rough drafts. You must complete the peer reviews using the supplied rubric **AND** make a comment on the overall paper no later than the Tuesday following the rough draft due date to earn credit on rough drafts. A copy of your lab notebook will earn zero (0) points regardless of completion of peer reviews. During the following lecture period, you will have the opportunity to discuss with your lab partners to make improvements to your final drafts.

After completing peer reviews and discussion with your lab partners, 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. Turning in an identical copy of the rough draft will result in zero (0) points on the final draft.

You are expected to make the recommended alterations to each report section as well as include all laboratories and an abstract for your **final manuscript**, which is **due April 24, 2023**. 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.

Tentative lecture schedule:

<i>Date</i>	<i>Discussion Topics</i>	<i>Assignment</i>	<i>Due Date</i>
January 11	Project overview Introduction Section PowerPoint	Syllabus Quiz	January 12 @ 11:59 pm
January 18	Introduction rough draft – peer edits	Lab Overview Quiz	January 19 @ 11:59 pm
January 25	Method Section PowerPoint	PCR primers Problem Set Lab 2 data set	Pre-lab 3 January 26 @ 11:59 pm
February 1	Methods rough draft – peer edits	---	---
February 8	Methods rough draft – peer edits	Restriction digest Problem Set 1	N/A
February 15	Results Section PowerPoint	Restriction digest Problem Set 2	Pre-lab 6
February 22	Results rough draft – peer edits	---	---
March 1	Results rough draft – peer edits	Protein expression Problem Set	Pre-lab 8
March 8	Discussion Section PowerPoint	Linear regions data analysis	March 21 @ 7:00 pm
March 22	---	Purification table	March 22 @ 7:00 pm
March 29	Discussion rough draft – peer edits	---	---
April 5	Abstract Section PowerPoint	Kinetics data analysis	April 5 @ 11:59 pm

Late Assignments

Laboratory notebooks will be kept on OneNote and locked the no earlier than the Saturday one (1) week following the lab. Since it is not “turned in” it cannot be turned in late. Please keep up with your notebook weekly.

Pre-lab 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 peer reviewed only. Instructors will not grade rough drafts. Full credit is earned on rough drafts by **turning it in on time AND completing peer reviews on time**. 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 ANY credit. A copy of your lab notebook will earn zero (0) points regardless of completion of peer reviews. **Failure to complete two (2) peer reviews prior to the lecture after rough drafts are due will result in zero (0) points for the rough draft.** Late submissions will be accepted with

the deductions described below, however, it is the student's responsibility to notify the instructor that the rough draft was late in order to be assigned peer reviews.

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

10 % deduction: < 24 hours after due date

25 % 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

Assignment	Points
Laboratory notebooks	200
Pre-lab quizzes	100 total
Lab performance	50
Lecture participation	50
Lab report rough drafts	40 total
Lab report final drafts	60 total
Final manuscript	100

Note that extra credit is built into the course assignments

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

A: > 540 pts

B: 539.9 – 480 pts

C: 479.9 – 420 pts

D: 419.9 – 360 pts

E: 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. Scores that have been posted to Canvas for a minimum of one (1) week are final.

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.

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 Molecular Biology/Protein Biochemistry</i>	<i>Streptomyces Microbiology</i>
January 12	1	Polymerase Chain Reaction	Plate Soil Samples
January 19	2	DNA Agarose Gel, PCR Cleanup & Restriction Digest	Select <i>Streptomyces</i>
January 26	3	DNA Agarose Gel & Cleanup, DNA Quantitation	Re-streak <i>Streptomyces</i>
February 2	4	Ligate DNA; Transform TOP10 Cells with Ligation Reactions;	Prepare <i>Streptomyces</i> plate for Antibacterial Test
February 9	5	Miniprep Plasmid DNA; Restriction Digestion	Spot Bacterial Test Strains for Antibacterial Test

February 16	6	Restriction Digest Analysis; Transform B121(dE3) Cells with Recombinant Plasmid; Pilot-Scale TEM1 Expression Trials	Start <i>Streptomyces</i> Liquid Culture
February 23	7	SDS-PAGE Analysis of TEM1 Pilot-Scale Induction	Isolate Resin from Liquid Culture
March 2	8	TEM-1 Purification from BL21(dE3) cells	Extract Metabolites from Resin
March 9	9	Purification tests	N/A
March 11-19		Spring Break	
March 23	10	Inhibition assays	Kinetics w/TEM-1 & Antibacterial test
March 30	11	<i>Virtual Laboratory</i> : Exploring TEM1 structure with PyMol	