# CHM 4300L – Laboratory in Biochemistry and Molecular Biology

#### **Instructor**

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**Office hours:** Monday 12:00 - 1:00 PM (hyflex); Tuesday 2:00 - 3:00 PM (online only); Or by appointment Office hours are held via HyFlex; Only one, masked student per in person office hours allowed.

## **Laboratory Manual** (available at Target Copy)

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

# **Class Meetings**

Pre-laboratory synchronous lecture: Wednesday 10:40 – 11:30 AM; AND 0021 Laboratory: BLR3: Thursday 9:35 – 12:35 PM; LEI 200

B3R6: Thursday 12:50 – 3:50 PM; LEI 200

#### **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 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.

If you are experiencing COVID-19 symptoms (<u>Click here for guidance from the CDC on symptoms of coronavirus</u>), please use the UF Health screening system and follow the instructions on whether you are able to attend class. <u>Click here for UF Health guidance on what to do if you have been exposed to or are experiencing Covid-19 symptoms</u>.

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 their station 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, 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.

# **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 and/or data analysis. Your active participation in these small group sessions will determine your lecture participation score.

To allow flexibility and a safe environment for students and the instructor, lectures will be delivered via HyFlex technology. Students who choose to attend in person class are expected to wear masks and be fully vaccinated to ensure everyone's safety. A WiFi enabled device with Zoom and headset, (and built in mic), is required for the classroom. Synchronous Zoom sessions are available for anyone who cannot, or does not want to, attend in person lectures.

## **Assignments**

OneNote will be utilized for E- lab notebooks, which will be graded for accuracy and completeness. Lab 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.

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 **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 help edit your classmates' drafts of the manuscript during specified lecture days.

Manuscript Section	Labs to be included	Rough Draft Due Date	Final Draft Due Date
Introduction	N/A	January 11	January 18
Methods	Labs 1-4	January 25	February 8
Results	Labs 1-7	February 15	March 1
Discussion	Labs 1-9	March 15	March 29
Full Manuscript	Labs 1-11		April 12

<sup>\*</sup>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.

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 April 12, 2022. 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:

Date	Discussion Topics	Assignment	Due Date
January 5	Project overview Introduction Section PowerPoint	Lab overview Quiz	January 10 @ 11:59 pm
January 12	Introduction rough draft - peer edits		N/A
January 19	Method Section PowerPoint	PCR primers Problem Set	January 20
January 26	Methods rough draft - peer edits		N/A
February 2	Experimental overview Methods rough draft – peer edits	Restriction digest Problem Set 1	N/A
February 9	Results Section PowerPoint	Restriction digest Problem Set 2	February 17
February 16	Results rough draft - peer edits		N/A
February 23	Results rough draft – peer edits	Protein expression Problem Set	February 24
March 2	Discussion Section PowerPoint		N/A
March 9	Spring Break		
March 16	Discussion rough draft – peer edits	Lab 9 Linear regions of data	March 22 @ 5:00 pm
March 23	Discussion rough draft - peer edits	Purification table	March 23 @ 11:59 pm
March 30	Abstract Section PowerPoint	Kinetics data analysis	March 30 @ 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.

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 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

# Grading

Laboratory notebooks200 ptsQuizzes100 ptsLab performance50 ptsSynchronous lecture participation50 pts

Lab report sections 100 pts (25 pts each)

Full Manuscript (labs 1-11) 100 pts Due April 12

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.

#### **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

<sup>\*</sup>You almost never make up the points lost from turning it in late. I don't recommend doing it!

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 <a href="mailto:umatter@ufl.edu">umatter@ufl.edu</a> 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)**

Date	Lab	DNA Molecular Biology/Protein Biochemistry	Streptomyces Microbiology
January 13	1	Polymerase chain reaction	Plate soil samples
January 20	2	DNA agarose gel, PCR cleanup & restriction digest	Select Streptomyces
January 27	3	DNA agarose gel & cleanup, DNA quantitation	Re-streak Streptomyces
February 3	4	Ligate DNA Transform TOP10 cells with Ligation Reactions;	Prepare <i>Streptomyces</i> plate for Antibacterial Test
February 10	5	Miniprep Plasmid DNA; Restriction Digestion	Spot bacterial test strains for Antibacterial Test
February 17	6	Restriction Digest Analysis; Transform Bl21(dE3) Cells with Recombinant Plasmid; Pilot-Scale TEM1 Expression Trials	Start <i>Streptomyces</i> Liquid Culture; Create Frozen stock
February 24	7	Pilot-Scale TEM1 Expression Analysis	Isolate/wash Resin from Liquid Culture
March 3	8	TEM-1 Purification from BL21(dE3) cells	Extract Metabolites from Resin
March 10	NO LAB	Spring Break	
March 17	9	Purification tests	

March 24	10	Inhibition assays	Kinetics w/TEM-1 & Antibacterial test
March 31	11	Virtual PyMol & AutoDock	