

Biological Chemistry Laboratory

CHE 554, Spring 2014

Instructor: Dr. Anne-Frances Miller
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Web Site: <http://www.chem.uky.edu/courses/che554/>

Meeting times and course logistics

Section 1-Lab: Tuesday and Thursdays from 1:30 PM to 3:30 PM in CP-236
Section 1-Lectures: From 12:30 to 1:20 in CP-222 (Tuesday and/or Thursday) Time slot may be used for laboratory exercises when not devoted to a lecture.
Section 2-Lab: Tuesday and Thursdays from 3:30 PM to 5:20 PM in CP-236
Section 2-Lectures: From 12:30 to 1:20 in CP-222 (Tuesday and/or Thursday) Time slot may be used for laboratory exercises when not devoted to a lecture.
Office Hours: Please speak with me during any of our assigned Lab times, or email to schedule an appointment. Remember, I am here to help you with this class. If you need help, I expect you to tell me.
Final Exam: Thursday 8 May at 10:30 am in CP-222
Midterm Grades: Midterm grades will be posed in myUK by the deadline established in the Academic Calendar (<http://www.uky.edu/Registrar/AcademicCalendar.htm>).
Absences: require written notice >1 week in advance and written documentation demonstrating that the absence qualifies as an excused absence under University regulations.

Course Description:

An introductory biological chemistry laboratory course. Areas of experimentation will include spectroscopic methods, electrophoresis, chromatography, and isolation and characterization of biological macromolecules. Prereq: CHE 232 or CHE 550 or CHE 552, and a physical chemistry course at or above the 400 level or consent of the instructor.

Student Learning Outcomes

After completing this course, participants will be able to execute work-horse techniques of biological chemistry, enabling them to hit the ground running in a first job. Participants will be able to understand the data produced by these techniques, determine whether the experiment has functioned correctly, diagnose and correct failures. Participants will be able to design experiments implementing the techniques (including controls) and make strategic decisions as to how and when to implement the techniques. Participants will understand the theory associated with the methods covered and be critical users and consumers of the data produced by the experiments discussed. Participants will be able to maintain a laboratory notebook conforming to professional standards.

Required Materials

Additional: Lab coat, safety glasses (provided), USB memory stick, Lab notebook with duplicate pages. This class will use chapters from the textbook "Experimental Biochemistry" (3rd Edition), Switzer and Garrity, W. H. Freeman and Company, New York. as well as numerous protocols from manufacturers and the literature. Prof. Miller will make these available to you, in electronic form, in addition to the notes for all of the lectures. It

is recommend that you print out the instruction pages and have them with you in lab. They will be posted on the course web site at least 24 hours in advance of each lecture, or they will be provided in class.

Teaching Assistants (2014)

Ms. Jianing Wang jn.wang@uky.edu

Ms. Ting Wang twa222@uky.edu

Course Grading

All lab reports combined account for 70% of your final grade. Each of the two exams will count towards 10% of your final grade. The remaining 10% will reflect all measures of laboratory conduct. This includes safety issues, being prepared, being cooperative, being independent, professional orderly conduct etc. The instructor will also offer a 'bonus' assignment intended to enrich and complement course content, but execution of this assignment is not required.

A score $> 94\%$ will be an 'A', $94 > x > 85$ will be a 'B', $85 > x > 74$ will be a 'C', $74 > x > 65$ will be a 'D' and lower scores will constitute failing grades.

Course Assignments: Lab reports

There are a total of 9 lab reports due in this class. The value of each report is listed at the end of this syllabus. The lab report **is** the set of original laboratory notebook pages, pulled from the notebook. The intention is that nothing need be copied over. Duplicate pages are the student's personal copy of the work, which will hopefully prove to be a useful resource in the future. All computer outputs such as graphs, charts and printed photos should therefore be printed in duplicate so that one copy can be taped into a designated spot in the original sheet (to be turned in) and another can be taped into the duplicate sheet (to be retained). All figures and charts also need to be provided with captions to integrate them into the narrative. Lab reports are read in detail by Teaching Assistants, who will provide detailed commentary. Both are then reviewed by Prof. Miller for a final grade. Be sure that you understand the comments as these are provided to aid in improving the lab performance and reports. Information concerning how to write your reports will be provided separately.

Reports not turned in on time, unless for approved excused reasons, will be given a 10% grade reduction for each week they are late.

The pre-lab section of your report must be COMPLETE before the lab exercise begins. T.A.s have been instructed to annotate lab report to indicate pre-labs that were not complete at the beginning of the laboratory period. Students whose pre-labs are not complete at the beginning of the laboratory period may not be allowed to execute their work. T.A.s are not obligated to stay late for students who had not completed their pre-labs on time and students may not work in the laboratory without supervision of a T.A. or Prof. Miller.

Lab reports are due **before class starts** on the day that is 2 weeks after the date on which the experimental activities were completed in lab, except for the last two lab reports which are due on 2 May (last day of classes) by midnight. Pages are to be in order and **stapled**. During the semester, reports should be turned in by placing them in the 'in' box in lab. Reports due on 2 May are to be put in Prof. Miller's faculty mailbox, *on top* of other mail.

Course Policies

The student is responsible for all equipment they use. In other words, if you break it, you are responsible for replacing it. The student will also be responsible for obtaining a lab coat (non-flamable), a lab notebook with

duplicate pages, and a USB memory stick. Lab work cannot begin until lab safety agreements are signed and returned.

Lab coats and goggles **must** be worn in the lab at all times. You must wear proper footwear and clothes that cover your legs completely and come up high enough at the neck to fill the 'V' of the lab coat neck. No loose hair, jewelry or clothing. Do not wear clothing that can catch on furniture or equipment, hang into reagents or ice buckets, or is long or tight enough to impede your movements in any way. Avoid flammable fabrics. Cell phones need to be turned off except for pre-approved reasons. Anyone behaving in an unsafe or disrespectful manner in the lab will have to leave the lab and a grade of zero will be assigned on the given lab assignment. For much of the time we will be wearing lab gloves. These are in part to protect our easily-contaminated biological reagents, but they also play a vital role in protecting biological chemists from hazards such as pathogenic or drug-resistant bacteria, genetic material and viruses. Despite the apparent safety of the reagents we will be using, students are required to observe standard safe practices in sterility and lab hygiene. Do not use gloved hands to touch food, cosmetics, your face, hair, doorknobs, cell phones, ipads nothing you will later touch without gloves on.

Biological reagents do not smell strong or have alarming colours, but a very tiny amount, even a single molecule, has the capacity to be deadly. Keep your brain on the job and do not cut corners. Similarly, if you see any other person in lab compromising on safety, speak up.

Cheating and plagiarism in any form will not be tolerated, and will result in a grade of zero for the assignment in question as well as a one-letter grade penalty on the course, at minimum. Please note that cheating takes many forms (see below). You are not allowed to share data without permission from the T.A. or the instructor. When a colleague's data are used to augment your own data in cases of experiment failure, your original data still need to be recorded and the data provided by a colleague must be accompanied by a statement of credit and an explanation of why a colleague's data are being used. The T.A./instructor's initials need to be present to recognize that the sharing has been approved. (Experiments do fail, there will be instances in which we will provide data to students whose own data does not provide a path forward.) You are allowed to discuss your experiments with others, and to get tips from others or watch how they do something, but the reports must be 100% your own data, your own words, and your own answers and conclusions. Everyone must execute their experiments and reports themselves independently of others.

Cameras will be available for students to use to document experimental apparatus set-up and results. However students preferring to use their own camera are welcome to do so. Photo-documentation of procedures, data and analysis need to be taped into the lab report and should be present in both the official report that will be turned in as well as the copy that will be retained.

Attendance

Because this is a laboratory class, and because all experiments are necessary to satisfy the requirements for ACS accreditation for this course, all students will be expected to complete all assignments.

Attendance at official times is mandatory. We cannot provide TA supervision on an individual basis. After the first *unexcused* absence, each additional *unexcused* absence will result in a single letter reduction in your final grade. Anyone more than 10 minutes late to lab will be considered absent and will not be able to participate on that particular day. (Recall that some labs will utilize the lecture hour.) Because of the nature of biological chemistry experiments, they are difficult to make up on an individual basis.

Excused Absences

Students need to notify the professor of absences prior to class when possible. S.R. 5.2.4.2 defines the following as acceptable reasons for excused absences: (a) serious illness, (b) illness or death of family member, (c) University-related trips, (d) major religious holidays, and (e) other circumstances found to fit “reasonable cause for nonattendance” by the professor.

Students anticipating an absence for a major religious holiday are responsible for notifying the instructor in writing of anticipated absences due to their observance of such holidays no later than the last day in the semester to add a class. Information regarding dates of major religious holidays may be obtained through the religious liaison, Mr. Jake Karnes (859-257-2754).

Students are expected to withdraw from the class if more than 20% of the classes scheduled for the semester are missed (excused or unexcused) per university policy.

Verification of Absences

Students may be asked to verify their absences in order for them to be considered excused. Senate Rule 5.2.4.2 states that faculty have the right to request “appropriate verification” when students claim an excused absence because of illness or death in the family. Appropriate notification of absences due to university-related trips is required prior to the absence.

Academic Integrity

Per university policy, students shall not plagiarize, cheat, or falsify or misuse academic records. Students are expected to adhere to University policy on cheating and plagiarism in all courses. The minimum penalty for a first offense is a zero on the assignment on which the offense occurred and a one-letter drop in the letter grade. If the offense is considered severe or the student has other academic offenses on their record, more serious penalties, up to suspension from the university may be imposed.

Plagiarism and cheating are serious breaches of academic conduct. Each student is advised to become familiar with the various forms of academic dishonesty as explained in the Code of Student Rights and Responsibilities. Complete information can be found at the following website: <http://www.uky.edu/Ombud>. A plea of ignorance is not acceptable as a defense against the charge of academic dishonesty. It is important that you review this information as all ideas borrowed from others need to be properly credited.

Part II of *Student Rights and Responsibilities* (available online <http://www.uky.edu/StudentAffairs/Code/part2.html>) states that all academic work, written or otherwise, submitted by students to their instructors or other academic supervisors, is expected to be the result of their own

thought, research, or self-expression. In cases where students feel unsure about the question of plagiarism involving their own work, they are obliged to consult their instructors on the matter before submission.

When students submit work purporting to be their own, but which in any way borrows ideas, organization, wording or anything else from another source without appropriate acknowledgement of the fact, the students are guilty of plagiarism. Plagiarism includes reproducing someone else's work, whether it be a published article, chapter of a book, a paper from a friend or some file, or something similar to this. Plagiarism also includes the practice of employing or allowing another person to alter or revise the work which a student submits as his/her own, whoever that other person may be.

Students may discuss assignments among themselves or with an instructor or tutor, but when the actual work is done, it must be done by the student, and the student alone. When a student's assignment involves research in outside sources of information, the student must carefully acknowledge exactly what, where and how he/she employed them. If the words of someone else are used, the student must put quotation marks around the passage in question and add an appropriate indication of its origin. Making simple changes while leaving the organization, content and phraseology intact is plagiaristic. However, nothing in these Rules shall apply to those ideas which are so generally and freely circulated as to be a part of the public domain (Section 6.3.1).

Please note: Any assignment you turn in may be submitted to an electronic database to check for plagiarism.

Accommodations due to disability

If you have a documented disability that requires academic accommodations, please email your instructor as soon as possible but not later than the end of the second week of classes. In order to receive accommodations in this course, you must provide me with a Letter of Accommodation from the Disability Resource Center (Room 2, Alumni Gym, 257-2754, email address: jkarnes@email.uky.edu) for coordination of campus disability services available to students with disabilities.

Laboratory Exercises

| Number | Report | Points |
|---------------------------|--|--------|
| Laboratory Assignment #1 | Molecular modeling, homology search and prediction of properties | 10 |
| Laboratory Assignment #2 | Spectrophotometry (serial dilution and determination of protein ϵ_{280}) | 15 |
| Laboratory Assignment #3 | ELISA | 10 |
| Laboratory Assignment #4 | Affinity Chromatography, SDS PAGE | 10 |
| Laboratory Assignment #5a | Molecular Biology (isolate plasmid, digest, agarose electrophoresis) | 15 |
| Laboratory Assignment #5b | Site-directed mutagenesis (primers, PCR, transform, select) | 15 |
| Laboratory Assignment #7 | Enzyme Kinetics (& kinetic mechanism) | 15 |
| Laboratory Assignment #8 | Biological fuel cell | 10 |
| Total possible | | 100 |

Report Guidelines

In this course, the lab report IS the lab notebook. All required information will be hand written in your notebook, and your notebook will be graded as your lab report. The only exception is processed data, which will be printed out and taped into your notebook, and the molecular modelling exercise. Note that you need two copies of all data in your notebook: one to hand in and one to keep. This includes taped-in material.

Your TAs will evaluate the reports and provide commentary (guidance). Please discuss these with your T.A. if you are in any doubt as to what is required. Your instructor will also be glad to discuss the reports with you. Each of the sections below will be graded independently using a grading scale of 1 through 10, where 1-2 is 'inadequate', 3-4 is 'weak', 5-6 is 'acceptable', 7-8 is 'good', and 9-10 is 'excellent'. The Pre-lab will be worth 30% of the final report grade, the Lab 55%, and the Post-Lab 15%.

Poor safety practices can result in loss of up to 50% of the earned grade. The lab report grade will be decreased by up to 20% if apparatus in the lab locker is left in poor condition.

Pre-lab

The pre-lab is to be completed before the laboratory session in which the assignment is to be executed. The handout for the lab will be available on-line at least 2 days in advance, via the lab web site. The presiding TA will grade your pre-lab before you begin the lab exercise. The pre-lab must contain the following sections:

One-column portion

1. *Experiment Motivation and Summary*: An overview of the experiment that you will be doing and why. Begin with a one-sentence objective.
2. *Experimental Theory*: A detailed description of the theory behind the major new techniques to be used, as well as the rationale for the steps to be taken in executing the experiment: why each step is taken and what it accomplishes, in theory. You can do this any way you feel is most effective, including drawing cartoons.
3. *Expected Outcomes*: A summary of what you expect to observe (nature of the observation as well as values / trends anticipated if all goes in accordance with plan and theory). Keep in mind both the technology and the particular experiment.
4. *Flow chart of steps and samples*: A road-map of what you will be doing that provides an overview of the steps involved in the day's activities including the intermediates retained vs. discarded, and a table of the samples that will be generated and analyzed.

Two-column portion

5. *Execution Plan*: Working in the left-hand side of the page, write a bullet for each action you will take, in essence creating an instruction list for yourself to follow during the execution itself.

Provide a blank space on the right-hand side for observations, next to each action listed.

Wherever you will need to record a value, use the right-hand side of the page to create a table ready to accept the data you intend to collect.

Similarly, create a right-hand column space wherever you want to insert a photograph or plot.

This exercise of creating spaces for the results you want to collect constitutes the advanced planning that can help prevent disasters when you are writing up the report and realize you never measured a volume or absorbance that you now need in order to calculate some critical value.

Leave plenty of space, skipping lines between steps, in order to allow inclusion of substantive notes on

observations or corrections during the execution.

This exercise should result in a fill-in-the-blanks template for success. It will also enable you to make MUCH more efficient use of your time in lab because you will know what you need to do and in what order.

Make note of any reagent that is dangerous, note precautions to take and provide a URL or reference to corrective action / antidote.

Lab

You are expected to write observations and measured values in your lab notebook as you conduct the experiment (real-time). This includes changes to your planned protocol, data recorded, real-time observations and any notes. **Nothing** should be written on scratch paper, and nothing should be rewritten. I will be looking in particular for the following:

1. The writing in the right-hand column should confirm or correct the planned execution so it is clear what was actually done.
2. The writing in the right hand column should include all raw data
3. The right-hand column should also include observations of such useful items such as colour changes, volumes, precipitation or viscosity changes, anything that provides information as to the state of the sample and the changes expected or not expected in its condition.

Be ESPECIALLY sure to record any accidents or unintended actions, such as addition of a reagent twice, and the actual duration of the time intervals (eg. your execution plan calls for a 30 min. incubation but you actually allowed 33.5 min.)

Post-Lab

Return to one-column format

1. *Data Analysis*: All processed data, including legends for each figure. This will mostly be graphs.
2. Final results obtained.
3. Insights, thoughts and implications.
4. *Questions*: Answer any questions asked within the protocol of the textbook. Be sure to make it clear which questions you are answering.
5. *Conclusions*: All experimental conclusions. These should be numbered. Explain in your notebook how the data support each of your conclusions.

In 10 sentences or less, restate what you did, the conclusions, and the learning outcomes**. Note whether the observed results were consistent with expectations, and provide plausible hypotheses for differences between expectations and observations.