

Course Syllabus

Biomolecular Chemistry 504: Human Biochemistry Lab

Spring 2018

Course Description

Biomolecular Chemistry 504 is an advanced lab course that introduces students to research experiences. We will begin by extracting and purifying lactate dehydrogenase (LDH), a key enzyme in human metabolism, and fully characterize this enzyme during the first part of the course. In the second part of the course, student groups will introduce mutations and conduct experiments to investigate how this affects the function of LDH using the model organism *Saccharomyces cerevisiae*.

Throughout the course there will be an emphasis on critically reading and analyzing primary scientific literature. Students will have the opportunity to improve their writing and oral communication skills through seminar-style presentation.

We will meet Tuesdays and Thursdays from 8:50-11:50am. You should expect to spend 1-3 hours a week outside of class to prepare and complete assignments.

Course Objectives

By the end of this course, students should be able to:

- Carry out fundamental techniques used in molecular biology & biochemical research.
- Explain the theory of fundamental techniques.
- Analyze and interpret experimental data to determine the results of an experiment.
- Apply statistical analysis to determine significance of results.
- Design experiments to directly address a scientific question.
- Maintain a record of experiments and communicate results through written & oral reports.
- Critically analyze experiments from peer-reviewed scientific publications.
- Demonstrate the ability to work effectively in teams.

Instructors & Teaching Assistants

Our goal is to guide you through the learning process and support you along the way. We will provide you with lectures and quizzes to help you master the material, and we will provide timely feedback on assignments and your performance in class. Please ask questions! We are all happy to talk with you.

Instructor/TA	Email	Hours Available
Dr. Angela Kita	amkita@wisc.edu	M-Th 12-4p, F 8-12p
Dana Dahhan	dahhan@wisc.edu	

Attendance & Accommodations

Students are expected to attend all scheduled class periods. Late arrival can affect your performance as you may miss important information given at the beginning of class. It is also disruptive to the class, affecting other students' performance. Attendance points will be lost for tardiness.

This is a fast paced summer session and absences should be avoided if possible. If you know you will need to absent during the semester, please discuss this with Dr. Kita during the first week of class and we'll try to make arrangements. If you are ill and cannot attend lab, please notify Dr. Kita via email *before class* begins otherwise it will be considered an unexcused absence. If you are unable to complete work on the scheduled day, an excuse form must be filled out stating your reason. Please discuss this with Dr. Kita and make sure any arrangements are signed off on. Unexcused absences cannot be made up. In accordance

with University policy for a religious observance conflict, students *should give notice within the first two weeks* of class of the specific lab dates for which relief will be requested.

If you require accommodations for testing or other components of the class, please schedule a meeting with the lab instructors within the first week of class. We are happy to discuss your needs and come up with a plan together. Please bring any documentation (i.e. McBurney Visa).

Student Preparation

This is a fast moving lab class and it will be important that you arrive prepared. This means (actually) reading the introduction and protocol and completing any pre-lab activities before coming to lab. I highly recommend sketching or writing out what you will be doing in lab before you arrive so that you have a better understanding of the protocol. Any special instructions (i.e. safety, reagent disposal, etc.) not found in the manual will be given to you at the beginning of the laboratory period.

Academic Integrity

By enrolling in this course, each student assumes the responsibilities of an active participant in UW-Madison's community of scholars in which everyone's academic work and behavior are held to the highest academic integrity standards. Academic misconduct compromises the integrity of the university. Cheating, fabrication, plagiarism, unauthorized collaboration, and helping others commit these acts are examples of academic misconduct, which can result in disciplinary action. This includes but is not limited to failure on the assignment/course, disciplinary probation, or suspension. Substantial or repeated cases of misconduct will be forwarded to the Office of Student Conduct & Community Standards for additional review. For more information, refer to studentconduct.wiscweb.wisc.edu/academic-integrity/.

Grades

Grades will be based on your lab notebook, lab reports, quizzes, data presentations, and your attendance and participation. The course is out of 500 points and your grade will depend on what percentage you earn: A=91-100%, AB=89-90.9%, B=81-88.9%, BC=79-80.9%, C=70-78.9%, D=60-69.9%, F<60%. This course is not graded on a curve, so if everyone in the class earns >91% then everyone gets an A. Your grade for this course will be based on your work in the following categories:

Assessment	Points	Percentage
Quizzes (5 x 20)	100	20%
Lab notebook/pre-labs (5 x 5)	25	5%
Lab Reports (9 x 10)	90	18%
LDH Purification Report	35	7%
Mini-proposal	25	5%
Paper Presentation	75	15%
Final Poster Presentation	100	20%
Participation	50	10%
Total	500	100%

Quizzes

There will be 5 quizzes covering the material from the previous lab. These may include questions about the theory of fundamental techniques, calculations, analysis and interpretation of experimental results or topics covered during discussion. Quizzes will be begin at the start of class. Make-up quizzes for excused absences will be different from those given in class.

Lab Notebook (5%)

You must have a dedicated notebook for the lab. You may choose your own notebook, but please add page numbers for easy reference. Lab notebooks will be checked periodically for completeness. Below are some guidelines:

- **Table of contents:** Lab notebooks should have a table of contents on the first 1-2 pages, and each entry should have a title and include the date. Be sure to fill out your table of contents page, using page numbers to reference each entry.
- **Pre-lab:** Before coming to lab you should read your manual and complete any pre-lab work assigned. For each experiment, you should include a title, a brief summary of the purpose/goal of the experiment (what will you be doing that day and why?) and answers to any pre-lab questions listed in the manual. Make sure to complete this before coming to class so you are prepared to work.
- **During lab:** Record any solutions, mixtures, or dilutions you made to complete your experiment. Record any data you collect such as spectrophotometry readings, how you loaded samples on gels, distance migrated for protein separation, or volumes collected.
 - Be sure to note any deviations from the protocol as listed in the lab manual—for example, if you let a reaction run for longer than the amount of time stated in the lab manual be sure to record this information.
 - Make sure your data is easy to decipher—using clearly labeled tables will help with this. Record any observations helpful to explain your results, especially if something unexpected occurs.
 - Include a summary—what were your main findings? What was the result of any calculations you did? *Any graphs generated should be taped into your lab manual here.*
 - We recommend that you leave extra space for each experiment in case you need to go back to add more information or calculations.
 - You may use pen or pencil, but please write legibly. If you need to make a correction be sure to cross out the writing or calculations, but avoid completely covering errors in case you need to refer back to that information.

Lab Reports (18%)

Each week you will submit a lab report summarizing your results. These reports must be typed, saved as a .pdf file, and uploaded to Canvas by the due date. At the end of each lab protocol in your lab manual you will find directions on what to include in your report. You will summarize your data as figures with figure legends and provide interpretation in a discussion section. These will be compiled into a final report (LDH Purification and Analysis Report). Check the schedule and/or Canvas for details on when each assignment is due. Points will be deducted for late submissions.

LDH Purification Report (9%)

In this report you and your partner will compile your data from labs 2-7. This will be an opportunity for you to collaborate, improve the work you previously submitted, and analyze the work as a whole. The report will follow the structure of a scientific journal article. You will include an introduction, a results section, and a discussion. More information will be provided in class.

Mini-Proposal (5%)

With a partner, you will select a journal article to present to the class. Partners will submit a mini-proposal on the paper they'd like to present, receive feedback, then prepare a short (~10 min) presentation.

Paper Presentation (15%)

You and your mini-proposal partner will present your selected paper during the final week of class.

Final Poster Presentation (20%)

Together with your group (3-4 students), you will create a poster presentation at the end of the semester. During the last week of class we will have a poster session during which you will share and discuss your results with other groups, the instructors, and invited faculty from the department. More information will be provided later.

Participation (10%)

You will earn points towards your final grade for participation. Daily attendance and participation are crucial components for success in a laboratory class, thus points will be awarded based on general performance including preparedness, timeliness, collaboration, following instructions, and maintenance of your lab bench. Arriving after the start of class (8:50 am) counts as being late.

Note on group-work

There are several assignments that involve working with a partner or group. You are responsible for making an equal contribution to any work that is submitted with others, and we will ask for peer-evaluations at the middle and end of the course. If you feel there is an unequal division of work, or a problem with timely completion of work, please come talk to me (Dr. Kita) as soon as possible. If necessary, students will be asked to complete work individually.

Additional Objectives & Assessment for Graduate Students

To receive credit for this course, graduate students should demonstrate the ability to:

- Generate ideas for original research in your field of graduate study & apply biochemical methods to investigate specific research aims.
- Predict possible shortcomings of a proposed experimental design and suggest alternate approaches.

These additional objectives will be assessed through a written research proposal. You will submit two drafts, receive feedback on each, and submit a final revised proposal. Please set up a meeting with Dr. Kita for more information.

Course Schedule

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Week	Date	Class	Assignment Due
1	1/23	Introduction to course Student surveys Figure analysis activity Lecture: Spectrophotometry	
	1/25	Introduction to lab work & lab safety Lab 1: Pipetting & Spectrophotometry	
2	1/30	Seminar/Discussion Lecture: Protein purification & LDH Mini-proposal assigned	
	2/1	Lab 2: LDH Extraction from tissue LDH Introduction (literature search)	Lab 1 report due 5pm (Figure creation)
3	2/6	Quiz 1 (Pipetting/Spectrophotometry) Seminar/Discussion Lecture: Chromatography	Submit paper selection for review (Discussion Board)
	2/8	Lab 3: Ion-exchange Chromatography	Lab 2 report due 5pm (LDH Intro & project scheme)
4	2/13	Lecture: Affinity tags Work on Mini-Proposal	
	2/15	Lab 4: Affinity Chromatography	
5	2/20	Quiz 2 (Chromatography) Lecture: Specific Activity Pre-lab work: Create outline Create excel spreadsheet for data analysis	Mini-Proposal due at 5pm
	2/22	Lab 5: Total Protein & LDH Activity	Submit data on Canvas Lab 3/4 report due (5pm) (Ion-exchange & Affinity)
6	2/27	Lecture: SDS-PAGE & Native gel electrophoresis Specific activity data analysis	
	3/1	Lab 6: Native gel electrophoresis	Lab 5 report due 5pm (Total Protein & LDH Activity)

Day	Date	Class	Assignment Due
	3/6	Quiz 3 (Specific Activity) Lecture: Enzyme Kinetics Pre-lab: Create excel spreadsheet for data analysis	
7	3/8	Lab 7: Enzyme Kinetics	Submit data on Canvas Lab 6 report due 5pm (Native gel)
	3/13	Quiz 4 (Gel electrophoresis) Lecture: Protein Structure & PyMOL	
8	3/15	Lab 8: Molecular Visualization w/ PyMOL	Lab 7 report due 5pm (Enzyme kinetics)
	3/20	Lecture: Cloning & Sequencing (Select mutations)	
9	3/22	Lab 9: Cloning & Sequencing Work on sequencing assignment	Lab 8 report due 5pm (PyMOL)
	3/27		
	3/29	<i>Spring Break: No Class</i>	
	4/3	Quiz 5 (Enzyme kinetics) Lecture: Presentations & Posters Outline seminar presentation	
10	4/5	Lab 10: DNA Miniprep & Yeast Transformations	Lab 9 report due 5pm (Sequencing)
	4/10	Lab 11: Spot test for activity, save samples (2-day cultures started on Sunday) Lecture: Western blot & antibodies	
11	4/12	Lab 12: Enzyme activity assays	
	4/17	Lab 13: SDS-PAGE & Western blot (part I)	Work on presentations
12	4/19	Lab 13: Western blot (part II)	Work on presentations
	4/24	Lab 14: Repeat experiments as needed	Work on presentations
13	4/26	Work day	Work on presentations
	5/1	Mini-symposium: Day 1	Submit presentations by 2pm Monday 4/30
14	5/3	Mini-symposium: Day 2	

Report	Topic	Due	Points
Lab 1	Figure (NADH curve) + legend	2/1	10
Lab 2	LDH Intro paragraph + purification scheme outline	2/8	10
Lab 3/4	Figures for ion-exchange column and affinity column	2/22	10
Lab 5	Specific activity data (tabulated), Bradford assay standard curve, protein activity figures, legends for each	3/1	15
Lab 6	Native gel analysis	3/8	15
Lab 7	Enzyme kinetics data (tabulated), figures of MM/LB for each assay	3/15	10
Lab 8	PyMOL	3/22	10
Lab 9	Sequencing	4/5	10
Summary	Figures from labs 2-7 edited & compiled, discussion section added	5/12	35