

Chemistry 373 - Biochemistry Laboratory II Spring 2024 Syllabus

Instructors: Agnes Pecak (aorlof@luc.edu), Flanner Hall 428, (773)508-2883

Teaching Assistants: Nick Kaley (nkaley@luc.edu)
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Laboratory Sections:

Chem 373-001: Wed 8:30 AM – 12:20 PM in Flanner Hall 016 TA: Leslie

Chem 373-003: Wed 8:30 PM – 12:30 PM in Flanner Hall 002 TA: Nick

Chem 373-004: Wed 1:40 PM – 5:30 PM in Flanner Hall 016 TA: Nick

Discussion Sections:

Chem 373-005: Mon 9:20 AM – 10:10 AM in Flanner Hall - Room 7

Chem 373-006: Mon 10:25 AM – 11:15 AM in Flanner Hall - Room 7

Office Hours: You may contact your instructor during their regularly scheduled Office Hours: Wednesday 12:30 pm-1:30 pm in person and by appointment.

If you are unable to contact the instructor directly, or by voice or e-mail, you may leave a phone message with the Chemistry Departmental Office, (773) 508-3100.

Course Pre-requisite: CHEM 372

Course Description and Objectives: The purpose of this second biochemistry laboratory course is to continue research on ADP-Glucose Pyrophosphorylase from *Escherichia coli* and a mutant version of the protein. This course will focus on characterization of the protein and the mutant through kinetic and thermal assays. Each student will be working on a recombinant ADP-glucose pyrophosphorylase from *Escherichia coli* and one mutant, N124A.

The objectives of the course are to:

- learn the theoretical foundations for the method used
- understand the applicability of the biochemical methods
- observe and record data accurately
- learn to present data, results and conclusions
- develop the ability to think scientifically and evaluate information critically

In the second part of biochemistry laboratory, students will focus on characterizing ADP-Glucose Pyrophosphorylase and its mutant N124A. There is a moderate risk of facing challenges. Some of the pedagogical goals are inspired by Kuhn, M.L., Figueroa, C.M., Aleanzi, M., Olsen, K.W., Iglesias, A.A. and Ballicora, M.A. (2010) "Bi-national and interdisciplinary course in enzyme engineering" *Biochem.Mol.Biol.Educ.*

38:370-379. [<http://dx.doi.org/10.1002/bmb.20438>]

... "that students work on real scientific problems during the laboratory sessions rather than performing a series of well-established experiments. While this may lead to unexpected difficulties, it is extremely advantageous for the student to learn how to approach a problem in an actual research environment"

Student teams are expected to perform experiments during their normally scheduled laboratory session time. Unfortunately, no student will be allowed to do work outside of their laboratory section.

A weekly 50-minute discussion section will be used for the discussion of procedures, results, and conclusions. The discussion will be conducted as an open forum of questions and answers between students and the instructor. With the instructor's help, the students will compare the methods that they have found in the original literature and determine which methods are best suited for the lab. Upon the completion of the course, the students should draw conclusions and insights about the structure-function relationships of this enzyme.

Required Materials:

- **Safety glasses/goggles:** No student will be permitted to conduct research without eye protection. If you need to wear regular glasses, then you will need to put goggles on top of your glasses.
- **Lab coat:** The assays performed in this lab use a dye that will stain clothing. Wearing a lab provides an additional barrier in case of chemical spills.
- **Gloves:** Disposable, nitrile gloves will be provided in the lab and should be worn when performing all experiments.
- **Face masks:** Masks are optional for all members of the lab.
- **Laptop:** A device capable of using the full version of Microsoft Excel and connecting to the Wi-Fi is required in the lab so data can be analyzed as experimental assays are run.

Safety: Laboratory safety is everyone's responsibility. By registering for this course, students agree to abide by all the safety precautions, information and rules provided in lab. Additionally, appropriate clothing must be worn that minimizes potential chemical contact with skin. Shoes that adequately cover the entire foot are required. Sandals, open-toed shoes, perforated shoes, open-backed shoes are not acceptable. No skin can be exposed on your feet or legs, so clothing that covers the body from the waist down (including ankles) should be worn. Failure to dress appropriately results in a student's dismissal from lab.

Tentative order of experiments:

Lab 1: Check-in/Kinetics Prep

Labs 2 and 3: Kinetics on WT and mutant ADP-glucose pyrophosphorylase (AGPase)

Labs 4 and 5: WT and mutant Fructose 1,6-bisphosphate (FBP) curves

Labs 6, 7, and 8: WT and mutant Adenosine Triphosphate (ATP) curves

Labs 9: WT and mutant Adenosine 5'-monophosphate (AMP) curves

Labs 10 and 11: Protein Thermal Shift of WT and mutant AGPase using qPCR

Labs 12 and 13: WT and mutant ligand dissociation PTS assays

Lab 14: Final Paper Due

Attendance/Participation: Participation is mandatory for ALL labs. Experiments in this course will be performed individually. In case of an absence, students must contact their instructor within 24 hours to determine a course of action for missed lab work.

Lab Reports:

There will be 5 lab reports written during the semester. After completing a set of labs, each student must turn in the lab report at the start of the next lab period. Note: the results for labs 12 and 13 will tentatively be combined in the final research paper. See Sakai for most up-to-

date information on lab reports and due dates as the semester progresses.

Lab report should have the following sections:

I. Title

II. Objective: give a one-or-two sentence statement of the purpose of the experiment

III. Procedure: briefly describe the kinetics setup

IV. Results/Calculations: all equations, sample calculation(s), charts, figures, graphs etc. that can be used to effectively present your results.

V. Discussion/Conclusion: the analysis and interpretation of your results. What do results mean? How do they relate to the objective of the experiment?

The lab reports must be typed.

A two-point deduction will be applied for each 24-hour period that a lab report is turned in late. Since lab reports will be submitted via Sakai, the deadlines apply regardless of lab attendance. Reports are always due 1 week after completing the set of labs.

Additionally, there will be a 2 or 4 point deduction on a lab report for missing one of the lab sessions covered in that report if there is no valid reason for the absence.

Grade Allocation:

50% Lab Reports. We expect you to follow a particular format for your research records, which is illustrated in this syllabus. There will be five lab reports for the semester, and **due dates and submissions will be on Sakai.**

5% Laboratory Performance. The TA in consultation with the instructor will assess this score, which will be based on proper use of instrumentation, good laboratory and leadership skills, observation of safety techniques, and on-time attendance. You are expected to arrive to the laboratory each week on time and be prepared.

10% Excel sheets with Data & Analysis. It is essential this semester that you present your data clearly. Each experiment and/or kinetic trial needs to have a date, title, sample's name (ex. T79A), and sample's concentration. Also, each trial must have a list of what was added to each tube, the results, etc. This data will be uploaded on Sakai after each set of experiments.

15% Discussion Section. A lab discussion is different than a lecture discussion in that much of the relevant background, prelab, and data analysis information are primarily only discussed then. This allows enough time for everyone to perform the experimental work during the lab time. The discussion score will be determined by the student's preparation, participation, and performance on quizzes and assignments done in the weekly discussion class. *There are no make-ups for discussion absences.*

20% Final Paper. This will be the continuation and expansion of your work from biochemistry laboratory 1. **The final paper due date is Wednesday, April 24 by 4 PM.** *If the final papers are submitted late, a four-point deduction will be added for each 24 hour-period of tardiness.*

Class Grades:

	A = 100-88 %	A- = 87-83
B+ = 82-78 %	B = 77-73 %	B- = 72-68 %
C+ = 67-63 %	C = 62-58 %	C- = 57-53 %
D+ = 52-48 %	D = 47-40 %	F = Less than 40 %

Interactions with TAs: In order to increase the amount of individual assistance you receive in lab, Teaching Assistants will participate in delivering this course. If at any time during the semester, you have any questions or concerns about the behavior of your Teaching Assistant, please contact the Lab Instructor.

Academic Integrity: All students in this course are expected to have read and to abide by the demanding standard of personal honesty, drafted by the College of Arts & Sciences, which can be viewed at: <http://www.luc.edu/cas/advising/academicintegritystatement/>

A basic mission of a university is to search for and to communicate the truth as it is honestly perceived. A genuine learning community cannot exist unless this demanding standard is a fundamental tenet of the intellectual life of the community. Students of Loyola University Chicago are expected to know, to respect, and to practice this standard of personal honesty.

Academic dishonesty can take several forms, including, but not limited to cheating, plagiarism, copying another student's work, and submitting false documents.

Any instance of dishonesty (including those detailed on the website provided above) will be reported to The Chair of The Department of Chemistry & Biochemistry, who will decide what the next steps may be. The penalty may include a grade of zero for that assignment and/or failure of the course.

Health, Safety, and Well-Being On-Campus: Please be familiar with and adhere to all policies and protocols posted on the Campus Info & Resources site: <https://www.luc.edu/healthsafetyandwellbeing/campusinforesources/>

Course Repeat Rule: Effective with the Fall 2017 semester, students are allowed only THREE attempts to pass Chemistry courses with a C- or better grade. The three attempts include withdrawals (W). The Department advises that it is preferable to complete a course with a grade of C or C-, and to demonstrate growth in future coursework, than to withdraw from a course.

After the second attempt, the student must secure approval for a third attempt. Students must come to the Chemistry Department, fill out a permission to register form or print it from the Department of Chemistry & Biochemistry website: <https://www.luc.edu/chemistry/forms/> and personally meet and obtain a signature from either the Undergraduate Program Director, Assistant Chairperson, or Chairperson in Chemistry. A copy of this form is then taken to your Academic Advisor in Sullivan to secure final permission for the attempt.

Student Accommodations: The Student Accessibility Center (SAC), Sullivan Center (773-508-3700), <http://www.luc.edu/sac>, has the mission "to support, service, and empower Loyola University Chicago students with disabilities" and to "Partner with faculty and staff to provide opportunities for collaboration, professional development, personal growth, and staff

interaction, as they relate to students with disabilities.” Please direct all questions concerning accommodations of disabilities to the Student Accessibility Center. Academic accommodations afforded to students require documentation and review. The Student Accessibility Center will issue accommodation letters for registered students to present to their instructors: accommodations are not active until students present these letters to their instructors. If students’ accommodations involve attendance or deadlines, instructors and students will jointly complete and execute an Agreement Form articulating their terms. See <https://www.luc.edu/sac/faculty/facilitatingaccommodations/> for guidance about implementing various kinds of accommodations in a way that is appropriate to your class. The Student Accessibility Center stands ready to work with you.

Loyola University Absence Policy for Students in Co-Curricular Activities

(including ROTC): Students missing classes while representing Loyola University Chicago in an official capacity (e.g. intercollegiate athletics, debate team, model government organization) shall be allowed by the faculty member of record to make up any assignments and to receive notes or other written information distributed in the missed classes. Students should discuss with faculty the potential consequences of missing discussion or laboratory classes and the ways in which they can be remedied. Students must provide their instructors with proper documentation (i.e., “Athletic Competition & Travel Letter”) describing the reason for and date of the absence. This documentation must be signed by the appropriate faculty or staff member, and it must be provided as far in advance of the absence as possible. It is the responsibility of the student to make up any assignments.
(<https://www.luc.edu/athletheadvising/attendance.shtml>)

Accommodations for Religious Reasons: If you have observances of religious holidays that will cause you to miss class/lab or otherwise effect your performance in the class, you must alert the instructor within 10 calendar days of the first class meeting of the semester to request special accommodations, which will be handled on a case by case basis.

Recording of Zoom class meetings: In this class, software **may** be used to record live class discussions. As a student in this class, your participation in live class discussions will be recorded. These recordings will be made available only to students enrolled in the class, to assist those who cannot attend the live session or to serve as a resource for those who would like to review content that was presented. All recordings will become unavailable to students in the class when the course has concluded. Students will be required to turn on their cameras at the start of class. Students who have a need to participate via audio only must reach out to the instructor to request audio participation only without the video camera enabled. The use of all video recordings will be in keeping with the University Privacy Statement shown below.

Privacy Statement: Assuring privacy among faculty and students engaged in online and face-to-face instructional activities helps promote open and robust conversations and mitigates concerns that comments made within the context of the class will be shared beyond the classroom. As such, recordings of instructional activities occurring in online or face-to-face classes may be used solely for internal class purposes by the faculty member and students registered for the course, and only during the period in which the course is offered. Students will be informed of such recordings by a statement in the syllabus for the course in which they will be recorded. Instructors who wish to make subsequent use of recordings that

include student activity may do so only with informed written consent of the students involved or if all student activity is removed from the recording. Recordings including student activity initiated by the instructor may be retained by them only for individual use.