

Chemistry 372 - Biochemistry Laboratory I
Fall 2023 Syllabus
Loyola University Chicago



Instructor: Agnes Pecak

Teaching Assistants: Nick Kaley and Leslie Castillo

Laboratory sections: Wednesdays 8:20 AM- 12:20 PM or 1:40 PM- 5:40 PM
Flanner Hall - Room 2 or 016

Discussion sections: Mondays 10:25 AM - 11:15 AM Flanner Hall Room 7 and 11:30 AM -12:20 AM Fanner Hall Room 7

Prerequisite: C- or better in CHEM 212, 214, 370, and 222 or 226.

Description and Objectives: This laboratory course is designed to simulate a research experience and to teach modern techniques utilized in a biochemistry laboratory, particularly the ones related to protein expression and purification. The course theme involves a comparative investigation of the enzyme ADP-glucose pyrophosphorylase and its mutant from a bacterial source. Each two-student team will be working on two forms (wild type and a mutant) of a recombinant ADP-glucose pyrophosphorylase.

The objectives of the course are to:

1. Learn and perform the techniques of mutagenesis, a recombinant protein expression and purification
2. Characterize the protein that has been previously constructed with a tag for rapid purification.
3. Learn about how to improve on methodology published, literature search and presentation.

ADP-glucose pyrophosphorylase is an enzyme well studied, in which its purification has been described for a long time. Here in the course, we will try to improve it with modern techniques. There is a certain but moderate risk of facing challenges. The instructor and teaching assistants have a vast experience with the system to guide the students, but ultimately, we will experience the enthusiasm of research. That is finding new things. 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"

The laboratory is an open-architecture environment. Student teams are expected to perform experiments during their normally scheduled laboratory session time; however,

there will be opportunities to repeat certain procedures or experiments.

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. There will also be small group assignments, quizzes given in class. 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:

- 1) Safety glasses: No student will be permitted to conduct research without eye protection
- 2) Notebook (lab)
- 3) Laptop (discussion)
- 4) Lab Coat is optional

Attendance/Participation:

Participation is mandatory for ALL labs. Every student is expected to come to every lab.

Safety:

Laboratory safety is everyone's responsibility. By registering in this course you agree to abide by all of the safety precautions, information and rules provided to you in lab.

Appropriate clothing must be worn that minimizes potential chemical contact with your skin. Shoes that adequately cover the entire foot are required. Sandals, open-toes shoes, perforated shoes, open-backed shoes are not acceptable. No skin can be exposed on your feet or legs, so clothing that covers and protects your body from the waist down (including your ankles) should be worn.

Laboratory Experiments: All proposed experimental procedures will be discussed and approved by the lab instructor.

1. Check-in; solutions preparation ex. media, mutagenesis, plasmid transformation, DNA purification, agarose gel electrophoresis (4-5 weeks)
2. Protein expression and purification of recombinant ADP-glucose pyrophosphorylase (allow 3 weeks)
3. SDS-PAGE (allow 1 week)
4. Kinetics of ADP-glucose pyrophosphorylase (allow 2-3 weeks).

Teaching Assistants (TAs): In order to increase the amount of individual assistance you receive in lab, Teaching Assistants will participate in delivering this course. They will be helping you get good data and provide any help you need

throughout each lab. They will be responsible for grading your lab reports and notebooks. If at any time during the semester, you have any questions or concerns about the behavior of your Teaching Assistant, please contact the Instructor.

Notebooks:

Notebooks should have the following sections: date, title, objective, deviations from the procedure (if any), calculations and results (ex. printouts of images). Notebook will be checked on a weekly basis by Teaching Assistants.

Lab reports: After completing a certain set of labs, each student will be required to submit the lab report in Sakai at the start of next lab. Please see the tentative schedule posted in Sakai.

Lab report should have the following sections:

I. Title

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

III. Procedure: describe the steps of the experiment such as concentrations, techniques, instrumentation and so on. It should be sufficiently detailed that the other experienced researchers would be able to repeat the work and obtain comparable results.

The procedure needs to be written in research paper style

IV. Results/Calculations: observations, equations, calculations, charts, figures, graphs etc which can be used effectively to present results clearly.

V. Conclusion/Discussion: the analysis and interpretation of your results. What do results mean? How do they relate to the objective of the experiment? Was the outcome successful? Outline the main conclusion of the project

A list of 5 lab reports:

- 1) Media preparation (Lab 1), Mutagenesis Part I and II (Labs 1 and 2)
- 2) DNA purification, digestion (Lab 3) and Agarose Gel Electrophoresis (Lab 4)
- 3) Plasmid Transformation (Lab 5) Protein Expression (Lab 6) and Purification (Lab 7, 8)
- 4) SDS-PAGE (Lab 9)
- 5) Malachite Green Assay (Lab 10,11,12)

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 point deduction on a lab report for missing one of the lab sessions covered in that report if no valid reason for the absence

Final Paper: The paper will be written in the format of a scientific journal: abstract,

introduction, materials and methods, results, conclusion, and references. *A two-point deduction will be applied for each 24-hour period that the final paper is turned in late.*

Grade Allocation:

50% Lab reports. You are expected to follow a particular format for your research records, which is illustrated in this syllabus.

10% Notebooks and 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, safety, taking experimental notes and observation of safety techniques. You are expected to arrive to the laboratory on time and be prepared.

20% Discussion Section. The discussion score will be determined by the student's participation, assignments and performance on quizzes. *There are no make ups for quizzes.*

20% Final paper. This paper will build on the lab reports, and will compare kinetic data submitted by other teams. Students will be required to draw conclusions about protein function based upon an analysis of the collated data from some other teams. Due date will be announced and posted on Sakai.

If the final papers are submitted late, one-point deduction will be assessed for each day 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 %	

Office hours: Outside of class, you may contact Agnes Pecak during regularly scheduled Office Hours, Wednesdays 12:30-1:30 pm in person and by scheduled appointment. The office location, telephone number, and e-mail address are: Flanner Hall 428, (773) 508-2883 aorlof@luc.edu

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

Sakai: This site contains current information for syllabus, experiments and procedures and scores.

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/>

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. In case a violation is detected, the particular assignment may receive a grade of zero.

Regarding the use of Artificial Intelligence: our Provost has expressed to Let us all make sure we are learning and sharing best practices and not allowing AI to do the learning for us.” In this course, any work you submit for credit must represent your own ideas and understanding of the assigned material. If you are uncertain about any case where your use of AI may be in conflict with University or course standards, please see me to discuss your concerns.

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).

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:

<http://www.luc.edu/chemistry/forms/> and obtain a signature from 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.

Students with Disabilities: If you have any special needs, please let me know in the first week of classes. The university provides services for students with disabilities. Any student who would like to use any of these university services should contact the Services for Students with Disabilities (SSWD), Sullivan Center, (773) 508-3700. Further information is available at <http://www.luc.edu/sswd/>.

Loyola University Absence Policy for Students in Co-Curricular Activities:

Students missing classes while representing Loyola University Chicago in an official capacity (e.g. intercollective 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 lectures and the ways in which they can be remedied. Students must provide their instructors with proper documentation describing the reason for and date of absence.

This documentation must be signed by an 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. If the student misses an examination, the instructor is required to give the student the opportunity to take the examination at another time.

(<https://www.luc.edu/athletheadvising/attendance.shtml>)

SmartEvals: Towards the end of the course, the students will receive an email from the Office of Institutional Effectiveness reminding them to provide feedback on the

course. They will receive consistent reminders throughout the period when the evaluation is open, and the reminders will stop once they have completed the evaluation. The evaluation is completely anonymous. When the results are released, instructors and departments will not be able to tell which student provided the individual feedback. Because it is anonymous and the results are not released to faculty or departments until after grades have been submitted, the feedback will not impact a student's grade. The feedback is important so that the instructor can gain insight into how to improve their teaching and the department can learn how best to shape the curriculum.

