General Information Advanced Proteins course 138039

Winter 2020-21: Lectures (start from Dec 21, 2020) Mondays 9:00-11:00, Labs February 23, 2021 to March 18, 2020 (see note* below)

Teaching Staff:

Instructors:

Dr. Oded Kleifeld [Biology 433-4, Tel 077-8871453 <u>okleifeld@technion.ac.il</u>] For office hours and/or virtual office hours by Zoom please schedule appointment by e-mail

Dr. Yael Pazy Benhar [Emerson 1-TCSB, Tel 077-8871901 yaelpb@technion.ac.il] Support:

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Prerequisites:

Introduction to Biochemistry and Enzymology (134019), Molecular Biology (134082), Metabolic Pathways (134113), Biochemistry and Metabolism Lab (134114), Cell Biology (134128) Students from other Faculties and/or students that did their undergraduate studies on other institute should be approved by the Instructors prior to registration (please contact Oded at okleifeld@technion.ac.il)

Credits: 5 points

Location: MALAT and Biology buildings (specific information will be provided prior to the beginning of the course)

Study hours per week/Semester:

- 1. Lectures: 2 hours per week (will start after Dec 21, 2020 till the end on the semester)
- 2. <u>Labs</u>: The main part of the course will be held over a concentrated 2.5-week period that due to different constraints it might spread over 3 weeks. During this period which lectures, labs and tutorials will be held between 9:00 to 17:00 every day (2-3 days during that time might be longer). During this time the students will be required to submit few tasks that will require
- 3. <u>Home assignments</u>: up to 4 home assignments will be given prior to and during the lab part of the course

Of note: the lab part schedule might change following changes in the circumstances (e.g. COVID-19-related restrictions, etc.)

Course Goals and Description

Proteins are important and are an essential component of every living system. The aims of this course are two-fold. First, to bring all students to the "same page" in protein biology, as Biology graduate students have different scientific backgrounds. Second, to familiarize the students with the classical and the modern methods of protein purification, kinetic analysis and structure-function analysis. The students will learn these methodologies by experiencing an entire purification process of a relatively stable enzyme from an easily available source. The course will include lectures, hands-on lab and bioinformatics tutorials. The course is meant for graduate students at the beginning (i.e. first year) of their degree in biology. A maximum of 16 students will be accepted. The course will be held over a concentrated 2.5-week period*(due to different constraints we will need to do 2 breaks during the course so in total it might spread over 3 weeks), during which lectures, labs and tutorials will be held for a full day every day.

Learning Outcomes

Upon successful completion of this course, students should be able to:

- Grow lyse and extract proteins from cells
- Concentrate protein solution
- Fractionate proteins based on specific properties (charge, MW etc)
- Purify proteins by affinity purification
- Separate and analyze proteins by SDS-PAGE
- Optional : detection of proteins by immunoblot (Western blots)
- Perform basic mass spectrometry- based proteomics procedures: in-gel digestion, in solution digestion, sample desalting
- Data analysis of simple proteomic experiments
- Kinetic analysis of enzymes including measurement and calculation of kinetic parameters for different reaction conditions
- Study protein Thermostability assessment with DSF
- Perform basic protein crystallography procedures
- Visualize, analyze and generate images/information of published protein structural data
- Use and collect and analyze protein information using online databases and bioinformatic tools

Course Content/Topics

The course will involve protein purification, enzyme kinetics, enzyme inhibition, multi-substrate assays, UV/VIS spectroscopy, ultrafiltration and precipitation approaches; low pressure chromatography by ion-exchange, affinity columns and gel filtration; Immunoblot (optional), electrophoresis, basic mass spectrometry protein identification, protein thermostability, protein interactions, crystallization and structure determination and the relevant bioinformatics.

Assignments and Grading Procedures

Home Assignments that will be given throughout the course and will include: Prelab report, Protein databases and online tools, Protein structure analysis, Short presentation about Protein purification methods (might be in couples): 30%

Student Participation: 20% Final written report 30%

Final presentation/assignment (Oral or via Zoom) 20%

In the case of dramatic changes in the circumstances (e.g. COVID-19-related restrictions, etc.)

Tentative Schedule Lab part

Day1	Day 2	Day3	Day4	Day5	Day6
Opening Lecture for Lab part	Protein activity (lecture)	Biophyscial Methods (lecture)	Structural Biology 1 (lecture)	Structural Biology 2 (lecture)	Guest Lecture
Purification 1: Lysis	Purification 1: Dialysis	Purification 1: Proteomics 1	Purification 1: Proteomics 1+2	Purification 1: Crystalyzation+X-ray (groups)	Purification 2: lysis
Purification 1: choromatography	Purification 1: concentration, SDS- PAGE, activity and protein assays	Purification 1: Proteomics 2	Protein databases tutorial	Purification 1: MST (groups)	Purification 2: concentration, SDS- PAGE, activity and protein assays
Purification 1: dialysis	Purification 1: proteomics 1				

Day 7	Day8	Day9	Day10	Day11	Day12
Purification 2: Precipitation	Purification 2: Chromotography 1	Purification 2: Proteomics-1	Proteomics Data analysis	Purification 1: Kinetics	Purification 1: Kinetics
Purification 2: Fraction collection	Purification 2: Chromotography 2	Protein databases tutorial	Proteomics Data analysis - Hands on	Purification 2: Kinetics	Purification 2: Kinetics
Purification 2: concentration, SDS- PAGE, activity and protein assays	Purification 2: concentration, SDS- PAGE, activity and protein assays		Protein Visualization - Hands on	Immublot	Immublot

Course Requirements & Course Policies

Attendance in all classes and labs is mandatory. Class will start exactly on time, and students arriving late will not be admitted. All students are expected to participate actively, and will be graded on this active participation.