Structural Biophysics:
Advanced Methods for 3-D Structure Determination of Biomolecules

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Guest Lecturers: All guest lecturers participated in the Spring 2021 version of the course

Offered: Spring Semesters
Credits: 3
Prerequisites: Basic biochemistry knowledge

Course Description: This course is a survey of advanced techniques for structure determination of biomolecules. We will discuss: (i) physical basis of cryo-EM, NMR and X-ray crystallography. Special emphasis will be put on cryo-EM, (ii) other emerging methods including: SAXS, FXS, Neutron Diffraction, CLEM, cryo-ET and FIB –SEM, (ii) advantages and disadvantages of each technique (iii) specific examples of bio-molecular complexes whose 3-D structures have been determined using one of the above methods. Except of learning theory, students will have the opportunity to visit state-of the-art cryo-EM and NMR facilities, and determine a high-resolution cryo-EM structure using a high-performance computing Linux cluster.

Textbook and Readings: There will be no specific textbook for the course. Students will be provided with Canvass or referred to resources available online or through Rutgers Libraries.

Course-specific Learning Goals: Upon completion of this course, students will be able to: (i) Understand the theory and practice of modern methods for 3-D structure determination of biomolecules. (ii) Calculate a high-resolution cryo-EM structure using high-performance computing. (iii) Deliver a scientific presentation in front of the class.

Exams, Assignments, and Grading Policy: The exam covering a theoretical part of the course will count for 40% of the grade. The ability to calculate a cryo-EM structure will count for 30% of the grade. The student presentation will count for 30% of the grade. We will also take into account class participation, especially in the structure calculation and student presentation part of the course. There will be no final exam.

<u>Assessment Plan</u>

1) Understand the theory and practice of modern methods for 3-D structure determination of biomolecules.

Assessment: exam including multiple-choice and essay questions.

2) Calculate a high-resolution cryo-EM structure using high-performance computing.

Assessment: we will provide computational resources and clearly define goals that will have to be achieved by students at the end of each session concerned with calculations of a cryo-EM structure. We will compare students' results with previously calculated benchmarks. For example, at the beginning of class #21, students will have to be able to demonstrate a set of 3-D structures calculated using a script: relion 3-D classification.

3) Deliver a scientific presentation in front of the class.

Assessment: students will be instructed during class #16 how to prepare a PowerPoint presentation and reminded how to analyze and evaluate a primary research article. At the end of the class, we will distribute articles. Students will be presenting these articles in a PowerPoint format during classes #24-28. Presenters will be assessed based on: comprehension (did the student demonstrate clear and concise understanding for the subject during the presentation and the Q&A session?), spoken language skills (did the student use the correct terminology, did she/he speak clearly, did he/she stay on topic making each slide transition to the next with ease, was the presentation creative?), written language skills (was the text on the slides clearly expressed, were pictures big enough to see, did the text on the slides show literacy in the given subject area?).

Grading Rubric:

Scoring	**Outstanding (90-100%)**	**Good (75-89%)**	**Satisfactory (60-74%)**	**Unsatisfactory (0-59%)**
Theory exam **(40% of the final grade)**	90-100% score at the exam including multiple-choice and essay questions.	75-89% score at the exam including multiple-choice and essay questions.	60-74% score at the exam including multiple-choice and essay questions.	0-59% score at the exam including multiple-choice and essay questions.
Practical -structure calculation **(30% of the final grade)**	A high-resolution 3-D structure was obtained. The student understands all processing steps and is able to clearly describe the processing pipeline.	A high-resolution 3-D structure was obtained. The student performed all processing steps, but is not able discuss details of the pipeline.	A low resolution 3-D structure was calculated. The student does not understand the processing pipeline and is not aware of the mistakes, which were made during processing.	The student didn't perform recommended processing steps. Hence the structure was not calculated.
Presentation **(30% of the final grade)**	The student demonstrated clear and concise understanding of the subject during the presentation and the Q&A session, used the correct terminology, spoke clearly, stayed on topic making each slide transition to the next with ease, the text on the slides was clearly expressed, pictures were big enough to see, text on the slides show literacy in the given subject area.	The student demonstrated understanding of the subject during the presentation and the Q&A session, and was able describe the material included in slides at ease. However, slides were not well prepared. Pictures were too small. Slides were crowded and contained too much text.	The student demonstrated understanding of the subject during the presentation and was able address some of the questions during Q&A session. However, he presentation was not rehearsed, hence the student often diverged of topic and was not able to adequately describe the material presented on slides. Slides were not well prepared. Pictures were too small. Slides were crowded and contained too much text.	The student did not understand the subject. The presentation was chaotic, and the student was not able to adequately describe the material presented on slides. Slides were not well prepared. Pictures were too small. Slides were crowded and contained too much text.

Daily Syllabus:

Class	Topic	Reading
1	Introduction	
2	Introduction to structural biology by Loren Williams (Georgia Tech).	https://ww2.chemistry.gatech.edu/~lw26/index.html
3	Basic concepts in EM: electron waves, field emission guns, lenses, energy filters and direct detectors	Canvas Online resources: http://cryo-em-course.caltech.edu/videos
4	Fourier transforms, Contrast Transfer Function (CTF), phase and amplitude contrast, defocus – Guest Lecturer Megan Dilorio	Canvas Online resources: http://cryo-em-course.caltech.edu/videos
5	EM data collection, image processing and structure calculations by Arne Moeller (Max Planck Institute, Germany)	https://moeller-lab.com/ Canvas
6	Algorithms for cryo-EM structure calculation – Guest Lecturer – Jason Kaelber	Canvas Online resources: http://cryo-em-course.caltech.edu/videos
7	Correlative Light and Electron Microscopy (CLEM), cryo-Electron Tomography (cryo-ET) – Guest Lecturer – Wei Dai	Canvas Online resources: http://cryo-em-course.caltech.edu/videos
8	Virtual tour of the cryo-EM and NMR facilities in the IQB	
9	Basic Concepts in NMR: energy levels, the vector model, chemical shifts, spin-spin coupling, chemical exchange, spin relaxation – Guest Lecturer - Andy Nieuwkoop	Canvas Online resources: http://www-keeler.ch.cam.ac.uk/lectures “Nuclear Magnetic Resonance” by P.J. Hore, Oxford University Press
10	Multidimensional NMR – Guest Lecturer - Guy Montelione	Canvas
11	Force fields and algorithms for NMR structure determination – Guest Lecturer - David Case	Canvas
12	Basic concepts in X-ray crystallography: crystal growth, X-ray generators and synchrotrons, Bragg’s law, diffraction– Guest Lecturer – Cathy Lawson	Canvas
13	Phasing methods and structure determination in X-ray crystallography - Guest Lecturer – Ezra Peisach	Canvas
14	X-ray crystallography- Protein Data Bank - Guest Lecturer – Stephen Burley	Canvas
15	Emerging technologies: Small Angle X-ray Scattering (SAXS) by Barak Akabayov (Ben Gurion University, Israel)	https://akabayov-lab.org/ Canvas
16	Visual storytelling and preparing a scientific presentation – Guest Lecturer – Maria Voigt Distribution of articles for student presentations	Canvas
17	Exam	

18	Cryo-EM structure calculation with Scipion: importing movies, motion and CTF corrections	https://scipion-em.github.io/docs/docs/user/user-documentation.html Canvas Handout
19	Cryo-EM structure calculation: particle picking, extraction, screening and 2-D classification	Canvas Handout
20	Cryo-EM structure calculation: 2-D classification, creating an initial model	Canvas Handout
21	Cryo-EM structure calculation: 3-D classification	Canvas Handout
22	Cryo-EM structure calculation: 3-D refinements	Canvas Handout
23	Fitting atomic coordinates to cryo-EM map with Chimera	https://www.cgl.ucsf.edu/Outreach/Tutorials/GettingStarted.html Canvas Handout
24	Student presentations and discussion	
25	Student presentations and discussion	
26	Student presentations and discussion	
27	Student presentations and discussion	
28	Closing discussion	

Academic Integrity

The university's policy on Academic Integrity is available at http://academicintegrity.rutgers.edu/. The principles of academic integrity require that all students:

- Properly acknowledge and cite all use of the ideas, results, or words of others.
- Properly acknowledge all contributors to a given piece of work.
- Make sure that all work submitted as his or her own in a course or other academic activity is produced without the aid of impermissible materials or impermissible collaboration.
- Obtain all data or results by ethical means and report them accurately without suppressing any results inconsistent with his or her interpretation or conclusions.
- Treat all other students in an ethical manner, respecting their integrity and right to pursue their educational goals without interference. This requires that a student neither facilitate academic dishonesty by others nor obstruct their academic progress.
- Uphold the canons of the ethical or professional code of the profession for which he or she is preparing.

Adherence to these principles is necessary in order to ensure that

- Everyone is given proper credit for his or her ideas, words, results, and other scholarly accomplishments.
- All student work is fairly evaluated and no student has an inappropriate advantage over others.
- The academic and ethical development of all students is fostered.
- The reputation of the University for integrity in its teaching, research, and scholarship is

maintained and enhanced.

Failure to uphold these principles of academic integrity threatens both the reputation of the University and the value of the degrees awarded to its students. Every member of the University community therefore bears a responsibility for ensuring that the highest standards of academic integrity are upheld.

Penalties specifically for graduate students:
Two levels of violations are recognized by the Academic Integrity Policy–separable and nonseparable. For graduate students nearly all violations are considered separable. Sanctions for separable violations include, but are not limited to:

- A grade of XF (disciplinary F) for the course.
- Disciplinary probation.
- Dismissal from a departmental or school honors program.
- Denial of access to internships or research programs.
- Loss of appointment to academically-based positions.
- Loss of departmental/graduate program endorsements for internal and external fellowship support and employment opportunities.
- Removal of fellowship or assistantship support.
- Suspension for one or more semesters.
- Dismissal from a graduate or professional program.
- Permanent expulsion from the University with a permanent notation of disciplinary expulsion on the student's transcript.

In this course, we will take cheating very seriously. All suspected cases of cheating and plagiarism will be automatically referred to the student conduct office (http://academicintegrity.rutgers.edu), and we will recommend penalties appropriate to the gravity of the infraction.