Molecular Biophysics III – Spring, 2007

Course Numbers: School of Medicine MSMBPH 2003 (CRN 17763)
Arts & Sciences MOLBPH 2003 (CRN 13096)
Credits: 4
Title: Biomolecular Interactions and Dynamics
Time: Tue & Thu: 3-4:45 pm
Place: BST3, 1st floor seminar room (Rm. 1018) and Structural Biology Department labs
Course directors: Judith Klein-Seetharaman (Structural Biology, University of Pittsburgh and Language Technologies Institute, Carnegie Mellon University) and Sanford Leuba (Cell Biology and Physiology, University of Pittsburgh)
Course teaching assistant: Oznur Tastan (Carnegie Mellon University)
Prerequisites: No formal prerequisites. Students typically have taken Biophysics I. Knowledge of the basic principles of different biophysical techniques is helpful. For students lacking this knowledge, we will provide suitable reading materials.
Course Website (Overview): www.cs.cmu.edu/~judithks/MB3_07.html
Course Website for enrolled students and participating faculty:
https://courseweb.pitt.edu/
Required textbooks: none, but list of recommended reading will be made

Contact information:

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Location: Suite 2.26g, Hillman Cancer Center, 5117 Centre Avenue in Shadyside Hospital (one can take the 71A bus from Oakland to the corner of Cypress/Centre Avenue or take one of the Shadyside Shuttle buses ["Oakland/Shadyside" or "Bigelow/Shadyside"] leaving from the front of Scaife Hall).
Website: http://cbpmedia.cbp.pitt.edu/leuba/leubabio.html

Course description: The goal of this course is to bridge experimental biophysical approaches with biological questions related to biomolecular interactions and dynamics. The course will contain wet-lab experimental demos and real-data analysis as well as lecture, tutorial and journal review & student presentation components (see course structure, below). At the end of the course, students should be able to weigh different experimental approaches against each other to address a specific biological question. They should know when a specific technique is applicable and when it is not or only in a limited fashion, know what information is obtainable from a specific technique and what is not, and how these different techniques complement each other. They should have a basic understanding of some of the major types of questions that are being addressed in modern molecular biophysics and structural biology, how the current knowledge in each field has been obtained, and what are open questions. They should be able to come up *ad hoc* with reasonable suggestions for how these open questions could be addressed. This ability will be tested in the Molecular Biophysics and Structural Biology seminar series, where students taking this course are expected to actively engage in the discussions immediately following the seminars. Students are invited to go for lunch with the seminar speakers to continue these discussions. For an updated list of speakers and topics in the seminar series, please visit www.cs.cmu.edu/~judithks/SBSeminar.html

Course structure: The course will consist of the following components: (1) Brief reviews of biophysical methods with emphasis of the information content and applicability of each technique relevant to biomolecular interactions and dynamics by the course directors. (2) Overview lectures given by the students of the course covering model systems in which a large number of different biophysical techniques have been applied to address biological functions. These lectures are 30-45 minutes long. Each student will present once. For each of the student lectures, the course instructors will provide a list of reading materials, including review or book materials, as well as a list of key experimental papers. The student that is to present needs to synthesize the material to fit into a 30-45 minute interval. The first draft of the lecture is reviewed by the course instructors, and we expect 1-2 iterations to finalize the lecture. The student then takes this lecture and discusses it with an expert in the field. This expert may also be present at the date the student presents the material and will assist in the discussion if needed. The other students not presenting also need to go through the materials. Preparation of a list of questions is mandatory. The list of questions will be handed in after the lecture. The questions will be the basis for the discussion of the material by the entire group of class attendants of that day (other students, course instructors, experts). (3) Protocol and data analysis tutorials on some of the major experimental tools used to study biomolecular interactions and dynamics. (4) Wet-lab experiment demos. (5) Review sessions by the course directors.

Student homeworks are of two types, (1) data analysis of data collected during wet-lab demo experiments and (2) preparation of the student lectures and summaries that serve as lecture notes (i.e. students are expected to spend a large amount of time preparing the lectures thoroughly with close feed-back loops to course directors and experts). Finally, we may print a book collecting the student presentations with the summaries.

Tentative Syllabus:

Title
Course overview and rules
Lecture 1. Introduction, Overview of Course, Rules and Expectations
Non-graded assessment exam. The goal of this exam is not to test the students, but to have a baseline of knowledge prior to the beginning of the class. This serves to assess student progress and effectiveness of teaching methods. If you do not know the answer, please provide guesses (it's okay to make "wild guesses") and notes on why you cannot answer the question ("I have never heard the word…", "I remember that there was something about… but I don't remember …")
Homework: Read papers on sample student overview “conformational changes in membrane receptors” and prepare questions

Lecture 2. Overview of biomolecular interactions and dynamics, outline of course topics, discussion of mock exam results **Lecture 3.** Sample “student overview”: conformational changes in membrane receptors
Lecture 4. Review of questions on conformational changes in membrane receptors
Module I: Introduction - comparison of bulk and single molecule biophysical methods
Lecture 5. Review of biophysical methods: bulk methods
Lecture 6. Review of biophysical methods: single molecule methods
Lab demo 1. Crystallization of lysozyme and demo of X-ray facility
Lab tutorial 1. Protocol and data analysis tutorial of lysozyme diffraction data
Lecture 7. Tutorial on structure visualization tools (Rasmol, Chimera, Pymol, VMD, O)
Homework: analyze the lysozyme structure that was automatically generated from the raw X-ray data
Lab demo 2. Atomic force experiment of DNA molecules and chromatin fibers
Homework: Comparison of length scales of images from lab demo results 1 and 2
Lecture 8. Review of questions on complementarities of different biophysical approaches. How realistic is a crystal structure? Comparison of H/D exchange coupled with NMR versus coupled with mass spectrometry, etc. Discussion of time scales of motions in prep for modules II and III.
Module II: Conformational changes
Lecture 9. Protein Conformational Changes Fundamentals – Allostery, Time-Scales and Amplitudes. Bulk and Single molecule approaches to study them.
Lecture 10. Student overview 1: Conformational changes transmitting information and/or transporting small molecules (ion channels)
Lecture 11. Student overview 2: Conformational changes generating force (molecular motors such as kinesins)
Lecture 12. Review of questions on conformational changes in ion channels and molecular motors
Module III: Protein dynamics
Lecture 13. Overview of protein dynamics and methods to study and analyze them
Lecture 14. Protein dynamics by NMR: application to folded and unfolded proteins
Lab demo 3. NMR: Setup of NMR experiments in NMR facility,
Lab tutorial 3. Potocol and data analysis tutorial in analysis of protein dynamics measured by NMR
Homework: Analysis of HIV protease NMR relaxation data
Lecture 15. Review of questions on protein dynamics
Module IV. Protein ligand interactions
Lecture 16. Overview of protein ligand interactions and application to drug discovery
Lecture 17. Student overview 3: Protein phosphatase drugs
Lecture 18. DNA / RNA binding. Identification and modeling of step times and sizes
Lecture 19. Student overview 4: Virus packaging
Lecture 20. Review of questions on protein ligand interactions
Module V. Posttranslational modifications
Lecture 21. Types of posttranslational modifications and effects on structure and function
Lecture 22. Identification of proteins and their modifications by matching masses to databases
Lab demo 4. Identification of proteins by mass spectrometry
Lab tutorial 4. Identification of proteins by mass spectrometry
Homework: Mass spec data analysis
Lecture 23. Student overview 5. Signaling complexes and their regulation by phosphorylation.
Lecture 24. Review of questions on posttranslational modifications
Module II: Protein-protein interactions
Lecture 25. Overview of protein protein interactions and methods to study them
Lecture 26. Student overview 6: Virus assembly
Lab demo 5: Cryo-EM of virus assembly

**Lab tutorial 5: **Analaysis of cryo-EM data
Homework: Cryo-EM data analysis
Lecture 27. Student overview 7: Native oligomerization versus Non-native homo-oligomeric interactions. Native oligomers such as allosteric enzymes versus fibril formation in various misfolding diseases.
Lecture 28. Student overview 8: Thermodynamics and Dynamics of Protein-DNA Complexes
Lab demo and tutorial 6: Biacore
Lecture 29. Review of questions on protein protein interactions
Homework: Protein-protein interaction kinetics and affinity determinations from Biacore experiments
Module VI. Folding
Lecture 30. Overview of folding. Energetics, kinetics and models of protein folding
Lecture 31. Residual structure in the unfolded state
Lecture 32. Methods to study folding using lysozyme as model system
Lecture 33. Student overview 9: Membrane protein folding
Homework: Circular dichroism data analysis
Lecture 34. Student overview 10: RNA folding
Lecture 35. Solvent effects on folding and stability of proteins
Lecture 36. Student overview 11: structure and stability of halophilic proteins
Lab tutorial 2: Optical tweezer and AFM pulling experiments and data analysis
Homework: Quantitative analysis of force measurements
Lecture 37. Review of questions on folding
Dates for 1^{st}, 2^{nd} and Final exams. Account for spring break dates.