MCB 65
Physical Biochemistry: Understanding Macromolecular Machines

The course aims to develop fundamental concepts of biochemistry as they apply to macromolecules, including protein and nucleic acid structure, thermodynamics and kinetics, ligand interactions and chemical equilibria. The course will also emphasize how these concepts are used in studies of the structure and function of biological molecules, including examples from metabolism. In the weekly section, students will undertake a discovery-based laboratory research project in which they will apply these concepts toward understanding the structure and function of the ATPase domain from the ABC transporter associated with antigen processing (TAP).

Prerequisite: (LPS A or LS 1a) and (Chemistry 17 or 20 or S-17 or S-20AB), Math 1b or higher is recommended

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CPM/Preceptor:	Rebecca LaCroix Office: SC 405	rlacroix@g.harvard.edu Office hours: Fridays 12-1 PM
Teaching Fellows:	Liam Theveny	ltheveny@g.harvard.edu Office hour: 5:30-6:30 PM in section room

Lectures: Monday, Wednesday, and Friday 10:30-11:45 AM EST; Biolabs 1087

Section: One mandatory combined discussion/laboratory session per week (synchronous) on Mondays, 3-5:30 PM EST

Grading:	Pop questions (best 10 of 12):	5%	
	Lab and Problem Sets (10):	30%	
	Final Lab Report:	10%	
	Midterms (3):	30%	
	Final:	25%	*****See alternative scheme on p. 3**

Textbook:
The Molecules of Life: Physical and Chemical Principles
By John Kuriyan, Boyana Konforti and David Wemmer
Garland Publishing, 2013

See additional readings on page 4 below.

Required Hardware:
To complete assignments, you'll need a <u>desktop or laptop</u> that meets the minimum requirements, and a <u>three-button mouse</u>. If you would like to borrow a Harvard-owned loaner laptop for this course, reach out to HUIT.

Academic Integrity Policy:
All work submitted by a student in MCB65 must be their own. Contributions from anyone or anything else must be properly quoted and cited each time that source is used. We specifically forbid the use of ChatGPT or any other generative artificial intelligence (AI) tools for any course assignment or assessment and at all stages of the work process, including preliminary ones. Use of generative AI or failure to cite sources constitutes a violation of the academic integrity policy of this course and the Harvard College Honor Code.

MCB 65 **Course schedule**				**Spring 2024**
Date	**Topic**	**Reading**	**Lecturer**	**LPS due**
M 1/22/24	Lecture 1 - Space, Energy and Time	Ch. 1A	Brewster	
	No Section			
W 1/24/24	Lecture 2 - Protein structure - the building blocks	Ch. 1.10-15	Brewster	
F 1/26/24	Lecture 3 - Protein structure - defining the elements	Ch. 4A,B	Brewster	
M 1/29/24	Lecture 4 - Protein structure shapes function	Ch. 4C	Brewster	1
	Section 1 – Journal Club			
W 1/30/24	Lecture 5 - Nucleic acid structure - beyond base pairs	Ch. 1.5-9, 2B	Brewster	
F 2/2/24	Lecture 6 - Anfinsen experiment - sequence determines shape	Ch. 5A	Brewster	
M 2/5/24	Lecture 7 - Sequence similarity scores - conservation and divergence	Ch. 5.4-15	Brewster	
	Section 2 – Short Quiz - PyMOL and mutant selection			
W 2/7/24	Lecture 8 - Protein structure - sequence-structure relationships	Ch. 5.16-end	Brewster	2
F 2/9/24	Lecture 9 - Conservation of energy - enthalpy and heat capacity	Ch. 6A	Prigozhin	
M 2/12/24	Lecture 10 - Energy and Force	Ch. 6B	LaCroix	
	Section 3 – Protein purification			
W 2/14/24	Lecture 11 - Macromolecular structure - forces at play	Ch. 6C to 6.21	LaCroix	3
F 2/16/24	Lecture 12 - Molecular Dynamics - using energy to calculate motions	Ch 6.22-23,Ref (1)	Prigozhin	
M 2/19/24	No lecture - President’s Day			
	No section			
W 2/21/24	**Midterm 1 - covers lectures 1-12**			
F 2/23/24	Lecture 13 - Multiplicity and Entropy I	Ch. 7.1-7.19	Prigozhin	
M 2/26/24	Lecture 14 - Multiplicity and Entropy II	Ch. 7.20-end	Prigozhin	
	Section 4 – SDS-PAGE analysis			
W 2/28/24	Lecture 15 - Determining macromolecule structures I	Ref (2)	Gaudet	4
F 3/1/24	Lecture 16 - Determining macromolecule structures II	Ref (2)	Brewster	
M 3/4/24	Lecture 17 - Energy Distribution and Entropy	Ch. 8	Prigozhin	
	Section 5 - Dialysis			
W 3/6/24	Lecture 18 - Boltzmann distribution	Ch. 8	Prigozhin	5
F 3/8/24	Lecture 19 - Defining free energy	Ch. 9	Prigozhin	
3/9-17/24	Spring Break			
M 3/18/24	Lecture 20 - Chemical Potential	Ch. 10A-B	Prigozhin	
	Section 6 – Protein concentration measurement			
W 3/20/24	Lecture 21 - Applying energy concepts to the protein folding problem	Ch 10.17-end	Prigozhin	6
F 3/22/24	Lecture 22 - Ligand binding I - Energetics	Ch. 12A	Prigozhin	
M 3/25/24	Lecture 23 - Ligand binding II - Case studies	Ch. 12B	Brewster	
	Section 7 – Protein stability assay			
W 3/27/24	Lecture 24 - Protein-protein interactions	Ch. 13B, Ref(3)	Brewster	7
F 3/29/24	Lecture 25 - Allostery	Ch. 14.7-14.20	Brewster	
M 4/1/24	Lecture 26 - Reaction kinetics	Ch. 15.1-13	LaCroix	
	Section 8 – Midterm exam review during section times			
W 4/3/24	**Midterm 2 - covers lectures 13-24**			
F 4/5/24	Lecture 27 – Reaction kinetics to enzyme kinetics	Ch. 15.14-end	LaCroix	
M 4/8/24	Lecture 28 - Enzyme kinetics - Michaelis-Menten and beyond	Ch. 16A,B	LaCroix	
	Section 9 – ATPase assay I			
W 4/10/24	Lecture 29 - Enzyme catalysis - examples	Ch. 16C	Brewster	8
F 4/12/24	Lecture 30 - Diffusion I	Ch. 17.1-20	Prigozhin	
M 4/15/24	Lecture 31 - Diffusion II	Ch. 17.21-end	Prigozhin	

	Section 10 – ATPase assay II			
W 4/17/24	Lecture 32 - Biological membranes to membrane proteins	Ch. 3B to 3.19	Brewster	9
F 4/19/24	Lecture 33 - Membrane proteins structure and function	Ch. 4D-end, Ref(4)	Brewster	
M 4/22/24	Lecture 34 – Metabolic pathways	Ref(5)	Brewster	
	Final exam review during section times			
W 4/24/24	Lecture 35 - Tuning functional modules of cellular functions	Ch. 14A, 14.21-22, Ref (6)	LaCroix	10
Tu 4/30/24	Final lab report due (reading period)			
TBA	**FINAL EXAM (Time & Location TBD)**			

For ALL course work you should show all of your work and justify your answers unless stated otherwise.

Pop Questions (12; counting for 5% of the final grade)
Pop Questions will be handed out *only* in class, *only* to students in attendance. Responses ***must be submitted on Canvas before the next lecture, unless otherwise instructed***. There will be 12 Pop Questions distributed through the semester. Your top 10 grades for the Pop Questions will be used to calculate the final grade (i.e. you get 2 free passes). Full credit for all these questions corresponds to 5% of the grade, 0.5% per question. There will be **no makeup**. Any student caught cheating will forfeit any Pop Question credit for the whole semester. You are allowed to use references and to discuss Pop questions with other students, but you should submit the answer in your own words. These questions are designed to test and further your understanding of some of the fundamental concepts discussed in class.

Laboratory and Problem Set Assignments (30% of the final grade)
You will complete ten combined LPS (Lab and PSet) assignments during the semester, each counting for 3% of your final grade. LPS assignments are due prior to the beginning of lecture (10:30 AM) on Wednesdays – **except for LPS1, which is due at the beginning of lecture on Monday, 1/29/24**. Late assignments will not be accepted. Answers to each problem set will be available on the web shortly after the due date. The LPS assignments are an integral part of this course, allowing you to apply the concepts covered in class to scientific questions and problems, including those which are relevant to the semester-long original research project you will conduct as part of section. Solving these problems will help you internalize the concepts and are a key preparation tool for the exams. The lab portion of the LPS assignments is designed to allow you to complete parts of a lab report throughout the semester and to receive feedback on your scientific writing prior to submission of your final report. You are encouraged to discuss the problems and lab-related writing in study groups, but the work you hand in must be your own writing and should reflect your thought process.

Lab Report (10% of the final grade)
The 10-15 page lab report, due at the end of the semester, will be worth 10% of the final grade. The lab report is your opportunity to present your original findings on your research project. Many components of the lab report you will have completed beforehand as part of the LPS assignments. We encourage you to start assembling the lab report early to allot time for revision and feedback. As resources, writing tips as well as detailed lab report guidelines and a grading rubric are available on the course website.

Midterms (3% Short Quiz, 2 Midterms 13.5% each, therefore counting for 30% of the final grade)
A short ~15-min quiz on amino acids, nucleotides and their properties in Section 2 will be worth 3%. Two Midterm exams will be 75 minutes long during the regular class period, worth 16% each. There will be no make-up exams for midterms. Students with a documented medical excuse will have their remaining exam scores each account for a larger percentage of the course grade.

Final (25% of the final grade)
The final will encompass material from the whole semester, although the final 1/3 of the material will be more prominent. Questions will be designed to apply what you learned through the semester to new problems.

*****Alternative final grading scheme**
We will also calculate your grade with your lowest Midterm exam counting for only 8.5% of the total, and the Final exam counting for 30%. We will use the higher of the two possible final grades to determine your letter grade. This provides you an excellent chance to redeem yourself if you end up disappointed with one of your Midterm grades.

Sections

Section attendance is mandatory. Section time(s) will be chosen based on student preferences with priority given to those using MCB 65 to satisfy concentration requirements. If you cannot attend one of the Monday afternoon sections, you cannot take this course. In section, you will undertake a discovery-based laboratory research project in which you will apply concepts covered in lecture toward gaining novel insights into the structure and function of the ABC transporter associated with antigen processing (TAP). In addition to laboratory work, section periods will be used to work through practice problems, review lecture materials, reinforce and clarify important concepts, answer questions and revisit problem set questions.

Suggested textbooks for supplemental reading and/or problems (Available on reserve at Cabot Library)

Protein structure

C. Branden and J. Tooze (1999) Introduction to Protein Structure. Garland Publishing

G. A. Petsko and D. Ringe (2004) Protein Structure and Function. New Science Press Ltd

Physical chemistry

R. Chang (2005) Physical Chemistry for the Biosciences. University Science Books

K.A. Dill and S. Bromberg (2003) Molecular Driving Forces. Garland Publishing

References for additional readings:

References (1) Molecular Dynamics - Required reading

- Karplus, M., and Petsko, G. A. (1990) Molecular dynamics simulations in biology. Nature 347: 631-639.

References (2) Macromolecule structure determination - *Optional* reading

- X-ray - Wlodawer, A., Minor, W., Dauter, Z., and Jaskolski, M. (2008) Protein crystallography for non-crystallographers, or how to get the best (but not more) from published macromolecular structures. FEBS J 275: 1-21.
- NMR - Wider, G. (2000) Structure determination of biological macromolecules in solution using nuclear magnetic resonance spectroscopy. Biotechniques 29: 1278-1282, 1284-1290, 1292 passim.
- EM – Carroni, M., and Saibil H. R. (2016) Cryo electron microscopy to determine the structure of macromolecular complexes, Methods, 95: 78-85.

References (3) Protein-protein interactions - *Optional* reading

- Janin, J., Bahadur, R. P., and Chakrabarti, P. (2008) Protein-protein interaction and quaternary structure. Quarterly Reviews in Biophysics 41: 133–180.

References (4) Signaling across membranes - *Optional* reading

- Hendrickson, W. A. (2005) Transduction of biochemical signals across cell membranes. Quarterly Reviews of Biophysics 38: 321-30.

References (5) Metabolism

Required reading

- Freddolino, P. L., and Tavazoie S. (2012) The Dawn of Virtual Cell Biology. Cell 150: 248-250.

***Optional* reading**

- Karr, J. R., et al (2012) A Whole-Cell Computational Model Predicts Phenotype from Genotype. Cell 150: 389-401.
- Goncalves, E. et al (2013) Bridging the layers: towards integration of signal transduction, regulation and metabolism into mathematical models. Mol BioSys 9: 1576-1583.

References (6) Building functional modules of cellular functions

Required reading

- Kuriyan, J., and Eisenberg, D. (2007) The origin of protein interactions and allostery in colocalization. Nature 450: 983-990.

<u>Optional</u> **reading**

- Ferrell, J. E., Jr. (1999) Building a cellular switch: more lessons from a good egg. BioEssays 21, 866-870.
- Milo, R., Shen-Orr, S., Itzkovitz, S., Kashtan, N., Chklovskii, D., and Alon, U. (2002) Network Motifs: Simple Building Blocks of Complex Networks. Science 298: 824-827.