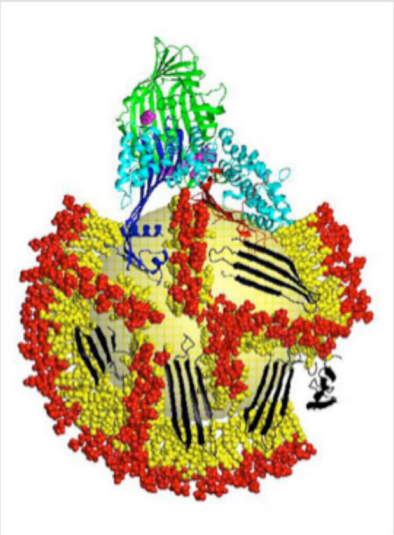
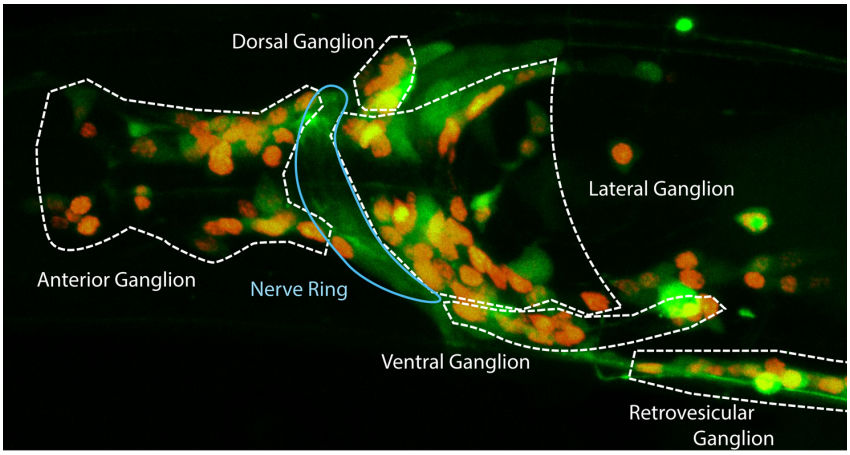
Graduate Programs in Physiology and in Biophysics (M.A./Ph.D.)



Lipoprotein model, cardiovascular research

Salamander rod, vision studies



C. elegans nervous system, neurophysiology studies

The Department of Physiology & Biophysics at Boston University School of Medicine

Table of Contents

Revised January 2021 (Bullitt/Atkinson)

Graduate Programs in Physiology and in Biophysics (M.A./Ph.D.)

I. Purpose and Background

The Department of Physiology & Biophysics brings 23 active faculty members together to provide excellence in research and graduate education. Research interests in the Department span the modern areas of Cellular Physiology and Molecular Biophysics, with strong concentrations in Structural Biology, Cellular and Neuro Physiology, Muscle Physiology, and the Structure and Biology of Lipids and Lipoproteins. Members of the faculty are nationally and internationally recognized as leaders in their individual areas of research. The Department provides flexible graduate programs with pathways leading towards a degree with a concentration either in Physiology or in Biophysics. The Department offers these two programs in a strong, collegial environment to encompass and promote the diverse overlapping research of all members of the Department.

The Department of Physiology & Biophysics graduate program is a component of the Boston University School of Medicine, Graduate Medical Sciences, umbrella Program in Biomedical Sciences (PiBS). The PiBS program integrates the foundations of interdisciplinary biomedical research with focused investigation in a specialized area together with preparation for career advancement. In the first year of the program, the students are undifferentiated and undertake a core curriculum in Foundations of Biomedical Sciences (FiBS), together with elective courses in area-specific core disciplines, and professional development. They engage in at least three eight week laboratory rotations, and select a faculty advisor. On completion of the first year, students join the department or program of their selected faculty advisor and proceed to fulfill the specific requirements of the departmental program.

The goal of the programs in Physiology & Biophysics is to produce graduate students who understand the thermodynamic, chemical, electrical and structural aspects of biological systems at the atomic level and in the context of the cell and organism. Students can elect a concentration in either Biophysics or Physiology. The training provides graduates the advantage of learning Physiology and Biophysics in the context of the regulation of homeostasis and pathogenicity of the cell, and of the organism as a whole. Graduates are trained to communicate and collaborate effectively with a broad range of disciplines ranging from medical personnel to chemists, engineers and physicists.

In addition to the courses in the FiBS curriculum, students interested in Physiology and Biophysics take a set of required core courses in the first year and second year that will lead to a level of understanding of the two disciplines necessary for a degree with a concentration in Physiology or in Biophysics. After completing the core course requirements, students have flexibility to choose the appropriate additional course work for their interests, within the guidelines set out below. Students who successfully complete the program are awarded either a Masters or Ph.D. in Physiology or in Biophysics depending on their concentration.

II. Administration of the Programs in Physiology and in Biophysics

The Chair of the Department of Physiology & Biophysics is the Director of the Graduate Programs and has the ultimate responsibility for administering the Graduate Programs in Physiology and in Biophysics. A Program

Steering Committee oversees the Graduate Programs and advises the Chair of the Student Affairs Committee (SAC) on specific needs of the Programs. The Steering Committee selects members of the faculty to serve on the SAC and oversees the appointment of committees that deal with developing new courses along with the review and updating of existing courses. The Steering Committee and the SAC Chair work jointly to supervise the appointment of faculty members to run the Departmental Seminar Series and plan the Departmental Retreat. Student Seminar Days are organized by the faculty members who are running the Department Seminar Series in a given year.

The Student Affairs Committee (SAC)

The SAC oversees the day-to-day operations of the Graduate Programs, including: orientation, assignment of dissertation advisors and administering the qualifying examinations. The SAC is comprised of 5-7 faculty members that adequately represent the diverse research interests within the Department. The SAC serves many roles in the Department. Foremost is the recruitment, evaluation and acceptance of students into the Programs. The SAC also handles the orientation of new students into the Programs. The SAC is charged with administering the Qualifying Examination. The SAC is also available to help with student problems and to mediate issues between students and Advisors.

III. Admission into the Programs in Physiology and in Biophysics

Student recruitment

The Programs in Biophysics and in Physiology seek students from a wide range of backgrounds including Physics, Chemistry, Biochemistry, Biology, and Medical Sciences. The Department has two representatives serving on the PiBS admissions committee that work to guide the acceptance of high quality students. For acceptance into the Department Program students should have outstanding grades in a rigorous curriculum and the program seeks applicants that have completed Organic Chemistry, Physics and Physical Chemistry courses.

The Ph.D. and M.D./Ph.D. Program

Requirements for Ph.D. and M.D./Ph.D. students are similar. Students in the Ph.D. Program take their qualifying examination in their second year. M.D./Ph.D. students enter after completing the second year Medical Curriculum and are therefore treated as post-masters students with a lower course and credit requirement (32 versus 64 total credits), and take their qualifying exam at the end of their first year in the Program. Other students entering into the programs with a Master's degree from within the USA are treated similarly and take their qualifying examination at the end of the first year. In both cases, post-Masters students are able to choose a suitable set of courses tailored to their backgrounds and research interests, with input from the SAC during the first year and from their Advisor in following years. Transfer students are handled on a case-by-case basis, but may be able to take the qualifying examination at the end of their first year.

The M.A. programs

Masters programs in Physiology and in Biophysics are offered. The M.A. degree requires 32 total credit hours (paid for by the student) and requires either a literature-based thesis or a short, laboratory based thesis with two readers from within the Department.

In some cases, a qualified M.A. student may proceed into the Ph.D. program. This decision is dependent on the student's application and acceptance into the PiBS umbrella program through the PiBS admissions process.

IV. The path of a graduate student

On joining the Department, students follow a path of requirements with specific course work dependent on their concentration in either Physiology or Biophysics. Students with a concentration Physiology are required to take at least one course in Biophysics and students with a concentration in Biophysics take at least one course in Physiology.

Special Topics Seminar Course (BY 871/2 and PH 841/2)

In the second and subsequent years, all students in the Department take the Special Topics Seminar course aimed at developing the student's ability to read the scientific literature and present the merits and/or deficits of a current research paper to other students and a proctoring faculty member. The students use a blackboard and computer projector during their presentations. This component of the course meets for 1 hour each week, as specified by the faculty member proctoring the class. Typically, all students present at least once each semester, and possibly more often, based on the number of enrolled students. Additionally, students are taught to write grants, research papers, and more general scientific articles such as 'News and Views' on papers from the current literature. An important complement to the student presentations is required attendance at the Physiology & Biophysics Seminar Series where the students are exposed to cutting edge research by outstanding speakers.

All graduate students in the department enroll in this course each semester for the duration of their degree program, earning 2 graded credits per semester.

The Faculty member who teaches this course is rotated after each semester, to allow a fresh viewpoint and area of expertise to be covered. The Chair of the SAC, with input from the Program Steering Committee, selects 6-8 Faculty who are interested in teaching this course on a rotating basis.

Written dissertation proposal

Success in science requires that students be able to express their thoughts both verbally and in written form. The necessary verbal skills are developed through participation in the Special Topics seminar course and by giving presentations in yearly research seminars at the Student Seminar Day(s), laboratory group meetings, pre-thesis meetings, and the dissertation defense seminar. Written skills are developed in required course work and in the writing of papers and the dissertation. However, the latter may occur rather late in a student's research project when time is at a premium. Thus, for their written Qualifying Exam, students are required to prepare an 8-10 page research proposal on their thesis project using the NIH National Research Service Award (NRSA) or American Heart Association Research Training Grants and Fellowship format and forms, as appropriate. This exercise begins with instruction, assignments, and feedback during the Special Topics course, with input from the advisor. The final research proposal allows the advisor and student to clarify the proposed research and provides the student with a forum to learn basic scientific and grant writing skills. This proposal should be completed during the Spring as part of the Qualifying Examination and is intended to help prepare the student for future writing of the dissertation, related papers, fellowships and grants. When the proposal is completed to the

satisfaction of the advisor, it is given to members of the pre-dissertation committee not less than two weeks prior to their first meeting, to serve as an introduction to the student's project. A copy of the proposal is also submitted to the SAC.

Student Seminars

In years 3-5, or starting with the Spring term after the Qualifying Examination has been completed, all graduate students present a 20-30 minute seminar on their research. These seminars form a logical extension of the Departmental Seminar Series and the Special Topics course and take place on a specified Student Seminar Day (or days) scheduled in April or May. Pre-dissertation committee members for a presenting student take notes on the quality of the presentation and meet with the student within 2-3 days, on either a one-to-one basis or in small groups with other committee members, to provide feedback with the goal of improving the student's presentation skills.

Student Posters Presentations

Students who have completed their Qualifying Exams present a poster during Russek Student Achievement Day in the Spring of each year, to improve their organization and presentation skills. These students also present their posters at the Departmental Retreat held each Fall. In addition, students are encouraged to enter their posters in the Science Day poster competition held at the Boston University Charles River Campus.

V. Course requirements

Students matriculated into the PiBS program follow a common curriculum in the first year covering both core disciplines and professional development. Formal coursework (total of 24 graded credit hours) is normally completed within the first two years of study, with the majority of courses taken during the first year. A total of 64 credits is required to fulfill the requirements of the Graduate School; credits accrued during a Masters degree can be considered for inclusion.

Foundations in Biomedical Sciences (FiBS) integrated core curriculum: PhD students are required to take at least 5 of the following two-credit courses, with modules 701, 702, 703, 704, and 707 mandatory; masters students are required to take at least modules 701,703, and 704:

*FC 701 – Protein Structure, Catalysis & Interaction
*FC 702 – Structure and Function of the Genome
*FC 703 – Architecture & Dynamics of the Cell
*FC 704 – Mechanisms of Cell Communication
*FC 707 – Physiology of Specialized Cells

FC 705 – Translational Genetics and Genomics
FC 706 – Molecular Metabolism

*To ensure rigor in data analysis, students are required to complete a statistics course relevant to their discipline

* Required

Professional Development: The two mandatory modules for PhD students are:

*FC 708 - Professional Development Skills

*FC 764 - Professional Presentation Skills

*Required

Courses in the Department of Physiology & Biophysics

The courses for first year students are chosen with the guidance of members of the both the PiBS and the Department's Student Affairs Committee (SAC), and in the second year with input from their Masters or Ph.D. Advisor.

Four modules of the university-wide Responsible Conduct of Research curriculum that includes initial online training and 4 in-person interactive workshops are taken by all students.

Ph.D. concentration in Biophysics: the course requirements include:

BY776/7 - Biophysics of Macromolecular Assemblies I and II,

BY762/3 - Foundations of Biophysics and Structural Biology I and II,

BY871/2 - Biophysics Seminar, and Special Topics

Ph.D. concentration in Physiology:

PH730 - Human Physiology A

PH731 - Human Physiology B

PH 841/2 Physiology Seminar and Special Topics – identical to BY871/2

Elective coursework is chosen jointly by the student and their advisor or thesis committee. To achieve a balanced curriculum for all students, the PhD degree requirements of the Division of Graduate Medical Sciences include a *minimum* of 24 graded credits in formal coursework. It is expected, although not required, that the formal course work credits are acquired prior to the qualifying examination at the end of the second year. Students who have already taken an appropriate graduate-level course in the core curriculum are permitted to substitute an elective or electives in the first semester

A sample curriculum

Fall semester, first year—all students

- Protein Structure, Catalysis, and Interaction (FC701, 3 cr)
- Structure and Function of the Genome (FC702, 3 cr)

- Macromolecular Assemblies I (BY776, 2 cr)
- Professional Presentation Skills (FC 764, 2 cr)

Spring semester, first year

- Architecture & Dynamics of the Cell (FC703, 3 cr)
- Mechanism of Cell Communication (FC704, 3 cr)
- Physiology of Specialized Cells (FC705, 3 cr)
- Foundations in Biophysics and Structural Biology I (BY762, 2 cr)
- Professional Development Skills (FC 708, 2 cr)

*Students fulfilling their physiology requirement with Human Physiology A and B would fulfill these requirements in their second year.

Remaining semesters, second year and beyond—all students

- Special Topics/Student Seminar (BY 871, 872, 2 cr each semester)
- Complete Course Requirements
- Electives

Suggested electives:

Department of Physiology & Biophysics

Cell Physiology (PH 843) 4 cr.

Human Physiology A (PH 542) 4 cr.

Human Physiology B (PH 543) 4 cr.

Foundations in Biophysics & Structural Biology I & II (BY 761 & BY 762) 2+2 cr.

Biophysics of Macromolecular Assemblies I & II (BY 776 & BY 777) 2+2 cr.

NMR Spectroscopy in Biology & Biochemistry (BY 772) 2 cr.

Metabolism & Cellular Function of Complex Lipids (BY 774) 2 cr.

Special Topics (PH 745, 746) 2 cr. (e.g. Biology of Vision, Calcium & Cell Function)

Courses in other Departments

Techniques in Biomedical Research (BI 777) 2 cr.

Comprehensive Immunology (MI 713) 4 cr.

Molecular Neurobiology and Pharmacology (PM 700) 4 cr.

Structure & Function of Proteins (BI 783) 2 cr.

Enzyme Catalysis (BI 788) 2 cr.

Receptors and Signal Transduction (BI 790) 2 cr.

Physical Biochemistry (BI 789) 2 cr.

Gene Regulation and Pharmacology (PM 880) 2 cr.

Mass Spectrometry, Proteomics and Functional Genomics (BI 793) 2 cr.

Principles of Genetics and Genomics (GE 701) 4 cr.

Biostatistics (MS 700) 2 cr.

Molecules to Molecular Therapeutics (MM 710) 4 cr.

Academic Performance

In accordance with the regulations of Graduate Medical Sciences, students will not receive course credit for grades below B-. Although B- is a passing grade for an individual GMS course, students must maintain a grade point average of B (GPA of 3.0) or higher. Students enrolled in the Ph.D. program are expected to maintain a GPA of B (3.0) or higher starting with their first semester and throughout their graduate career. Any student who fails to meet this standard is automatically placed on academic probation. The Student Affairs Committee (SAC) in the Department of Physiology & Biophysics or the Program in Biomedical Sciences Steering Committee will notify the student of his or her probation status by sending the student a notification letter or email that is copied to the student's academic advisor.

In order to remain enrolled in the program, a student on academic probation must increase his or her cumulative GPA to 3.0 in the next semester. If a student does not increase his or her GPA to 3.0 by the end of the semester after it fell below 3.0, the student will be dismissed from the program unless the student has petitioned the SAC and, due to extenuating circumstances, an extension has been granted. A student who has a GPA that falls below 3.0 at the end of the fourth semester will not be allowed to take the qualifying exams, and will have one additional semester to increase his or her GPA to 3.0 before dismissal from the program.

The Qualifying Examination for PhD Students

The Qualifying examination is given at the end of the second semester within the 2nd year for Ph.D. students. M.D./Ph.D., post-Masters and some transfer students in the Ph.D. Program have the option of taking the Qualifying Examination at the end of the second semester of the first year. Administration of the Qualifying Examination will be overseen by the SAC. Program Faculty members serve on the Qualifying Examination committees on a rotating basis. The two-part exam consists of:

1- For the written qualifying examination students write a research proposal that correlates with, but may or may not include his/her planned dissertation research. This document is judged by three members of the SAC committee for the student's understanding of the relevant scientific literature and ability to propose testable scientific hypotheses.

2- An oral examination where the student is assigned 2-3 current research papers to read and is subsequently tested on their understanding of the material in front of an examining committee. This committee is comprised of 5 Program Faculty members. Papers are chosen by the committee and given to the students 2-3 weeks in advance of the oral examination. Dissertation Advisors will not participate in the oral defense of students who are pursuing a Ph.D. in their laboratory.

Both the written and oral portion of the qualifying exam must be passed in order for a student to continue in the PhD. program. Students who fail either the oral or the written portion of the exam (or both) can retake that portion of the exam once in order to achieve a passing score. Masters students do not take the Qualifying Examination.

Pre-dissertation meetings

After the student has passed the Qualifying Exam, a pre-dissertation committee of at least five members must be established in the Fall of that year and submitted to the SAC. Students submit their dissertation proposal in grant form to the members of the committee two weeks before the first committee meeting. At least one member from outside the Department, and preferably from a different institution, should be included in the final dissertation committee. An external committee member is not required for pre-dissertation meetings, which will be held about every 10-12 months or ~3 times prior to graduation.

Dissertation defense

The Ph.D. thesis defense consists of a public seminar followed by a closed door thesis defense with a minimum of 5 committee members, including 1 member from outside the Department

VI. Resources for the Programs

All Faculty of the Department of Physiology and Biophysics participate in the Graduate Programs in Physiology and Biophysics. The Faculty have well-funded research programs and extensively equipped individual laboratories for carrying out research in Cellular Physiology and Biophysics. In addition, the Department maintains core facilities in Molecular Biology, Spectroscopy, X-ray crystallography, NMR and Structural

Electron Microscopy that may be used by students carrying out their research. The Faculty of the Department of Physiology and Biophysics and their research interests are listed below. A more in depth review of each Faculty members research can be obtained from the Department Web pages at http://www.bumc.bu.edu/phys-biophys .

Faculty Members Involved in Graduate Student Research	**Research Interests**
David Atkinson, Ph.D.	Plasma Lipoproteins: Structure & Biology
Christopher W. Akey, Ph.D.	Structural Biology of Channels and Chaperones
Esther Bullitt, Ph.D.	Bacterial Adhesion and Viral Replication
Lynne Coluccio, Ph.D.	Non-muscle myosin
Xiaohu Mei	High density lipoproteins
Christopher Connor, Ph.D.	Neuro-function of Anesthesiology
Christopher Gabel, Ph.D.	Neural Regeneration and Neurocircuitry in *C. elegans*
Olga Gursky, Ph.D.	Protein Structure, Function, Folding and Lipid Interactions
James A. Hamilton, Ph.D.	Structural Biology of Membranes in Native and Pathological States
Haya Herscovitz, Ph.D.	Assembly, Chaperone-Assisted Folding and Secretion of Lipoproteins
William J. Lehman, Ph.D.	Electron Microscopy and 3D Reconstruction of Actin Thin Filaments
Clint Makino, Ph.D.	Photoreceptor Physiology and Retinal Disease
Assen Marintchev, Ph.D.	Eukaryotic Translation Initiation and NMR
C. James McKnight, Ph.D.	Protein Structure, Function, Folding and NMR Spectroscopy
Raphael A. Zoeller, Ph.D.	Functional Roles of Lipids in Cell and Membrane Biology
Michael Rynkiewicz, Ph.D.	Muscle Function - x-ray crystallography and molecular dynamics
Hiroshi Tokuo, Ph.D.	Cytoskeletal dynamics and unconventional myosins

Appendix A1. Course syllabi (Examples)

Foundations of Structural Biology I: GMS BY 762
Structure Determination by Crystallography and Electron Microscopy

Course Director: Dr. David Atkinson, W308C, 8-8448, atkinson@bu.edu

Course format: The course will be conducted through a tutorial mechanism. Each week you will be given an assignment of topics to prepare for the following week's class. This preparation may require research on an underlying theory, analysis of the principles behind a method or learning and understanding the method and analysis used in a paper. You may be expected to watch and understand material presented in internet on-line course videos. You may need to use internet web resources and books in your preparation. You do not need to buy books since most are available from the faculty members. Working on topics as a group is encouraged.

Grading: Course grade will be determined though problem sets (2-3 per block) worked on independently. Participation in the course discussions will also contribute to grades.

10 September	Dr. Atkinson	**Introduction,** Perspectives and background of structural biology. Symmetry in structural biology
17 September	Dr. Atkinson	**Fourier Theory in Structural Biology** Waves, Fourier series. Fourier and inverse Fourier Transforms.
24 September	Dr. Atkinson	**X-ray Crystallography I** **Geometrical Diffraction:** Lattices, Unit Cells, Crystal Systems, Bragg's Law, Reciprocal Lattice, Space Groups, von Laue Conditions, Ewald sphere,
01 October	Dr. Atkinson	**X-ray Crystallography II** **Fourier Theory in Diffraction:** Importance in Structural methods, Convolution, Correlation
15 October	Dr. Atkinson	**X-ray Crystallography III** **Fourier Analysis of Scattering and Diffraction:** Atomic scattering, form factors, assemblies of atoms, lattices, electron density, Structure factor, Patterson function, resolution, phases and phase problem, symmetry and systematic absences.
22 October	Dr. Rynkiewicz	**X-ray Crystallography IV**

		Protein Crystallization: Preparing proteins; purification, concentrating, storage. Crystal growth: principles and methods, solubility, saturation, nucleation, batch, vapor diffusion and dialysis methods, micro and macro seeding, crystal storage and handling. Crystal soaking: cryo-protectant, heavy atoms, substrates, ligands or inhibitors.
29 October	Dr. Rynkiewicz	**X-ray Crystallography V** **Macromolecular Data Collection and Processing:** Data collection: Crystal mounting, radiation damage, cryo-techniques. Photography: Still, oscillation, precession, Laue, resolution, mosaicity. Data processing and reduction: Indexing, integration, error estimate, polarization correction Lorentz correction, absorption, space group determination, statistics.
05 November	Dr. Rynkiewicz	**X-ray Crystallography VI** **The Phase Problem:** Phase determination: Multiple isomorphous replacement, multiple anomalous dispersion, molecular replacement, direct methods. Phase improvement: Solvent flattening, histogram matching, non-crystallographic averaging.
12 November	Dr. Rynkiewicz	**X-ray Crystallography VII** **Model Building and Refinement:** Map calculation: Difference maps. Interpretation of electron density Maps: Model building. Refinement: Least squares, maximum likelihood, rigid body, group and individual B factor, positional, simulated annealing. Assessment: Conventional and free R-factors, real space correlation.
20 November	Dr. Bullitt	**Structural Electron Microscopy I** **Introduction and Electron Optics:** Introduction to Electron Microscopy and its use in Cell and Structural Biology Comparison of electron and light optics Phase contrast microscopy Contrast and image formation Recording images: CCD vs CMOS vs Direct electron detection
27 November	Dr. Bullitt	**Structural Electron Microscopy II** **Radiation Damage, Specimen Preparation and the Projection Theorem:** Radiation damage and biology: Minimal dose and low temperature Theory of specimen preparation for thin sections, negative staining and frozen-hydrated work The projection theorem and its application to 3D structural analysis of electron micrographs
04 December	Dr. Bullitt	**Structural Electron Microscopy III** **3-Dimensional Image Reconstruction, Overview and Single Particle Analysis (SPA):** Cross correlation and Bayesian alignment

		Common lines methods in reciprocal or real space Single Particle Analysis Classification methods Random conical tilt 3D reconstruction
11 December	Dr. Bullitt	**Structural Electron Microscopy IV** **Electron Tomography (ET); Correlative Light & Electron Microscopy (CLEM):** Dose fractionation Merging data to form a 3D reconstruction (back projection vs SIRT) CLEM using fluorescent labels, and CLEM/FIB (focused ion beam) for cryo-ET
18 December	Dr. Bullitt	**Structural Electron Microscopy V** **3D Reconstruction of Helical Filaments and 2D Crystals:** Helical symmetry and the Fourier transform of a helix Indexing helical diffraction patterns (near and far side 2D projections) 3-D helical reconstruction using Fourier-Bessel techniques or SPA

Foundations of Structural Biology II: GMS BY 763

Computation, Nuclear Magnetic Resonance, Thermodynamics and Spectroscopy in Structural Biology

Course Director: Dr. Atkinson, W302, 8-4015

16 January	Dr. Atkinson	**Introduction,** **Structural Databases and Tools:** Protein Data Bank (PDB), PDB structure files, EM Structure repository (EMDataBank)
23 January	Dr. Atkinson	**Structural Computational Biology I** **Macromolecular graphics, structure visualization and analysis.**
30 January	Dr. Atkinson	**Structural Computational Biology II** **Molecular Mechanics and Dynamics**
6 February	Dr. McKnight	**Structural NMR I** **Introduction to Fundamental Aspects of NMR:** Nuclear spin, Zeeman splitting, Boltzman distribution, precession of spins, Bloch equations, one pulse NMR experiment, spin relaxation, linewidth, chemical shifts, j-coupling, dipole-dipole interactions (NOE).
13 February	Dr. McKnight	**Structural NMR II** **Experimental Aspects of NMR:** Sample considerations and conditions. Instrumentation: Tour of an NMR spectrometer, data acquisition,

sampling theorem, quadrature detection, phasing, lock channel. Data processing: Fourier transforms, apodization, zerofilling, linear prediction, referencing, and integration. Water suppression: Presaturation, gradient, jump-return, spinlocks, solvent deconvolution.

20 February Dr. McKnight **Structural NMR III**

Multidimensional and Heteronuclear NMR: Through bond experiments: COSY and TOCSY. Through space experiments: NOESY and ROESY. Heteronuclear experiments: HMQC, HCCH-COSY. Combining experiments: HMQC-NOESY, HMQC-TOCSY. Three dimensional experiments: 3D-HMCQ-NOESY. Triple labeling: HNCA, HN(CO)CA . Deuterium labeling of large proteins.

27 February Dr. McKnight **Structural NMR IV**

Hydrogen Exchange and Relaxation Measurements: Hydrogen exchange rates and protections factors. Relationship of protections factors to ΔG. Pulsed HX and protein folding. Theory and mechanisms of relaxation (T1, T2, and NOE).The spectral density function. Experimental aspects to measure T1, T2 and NOE.

6 March Dr. McKnight **Structural NMR V**

Sequential Assignment and Structure Calculation: Proteins: Spin systems sequential NOEs, medium range NOEs, stereospecific assignments, direct methods with triple labeled samples. DNA: Spins systems, sequential NOEs. NMR distance, angle, and chemical shift restraints. Distance geometry, simulated annealing, and relaxation matrix back-calculation. Software packages. Judging the quality of NMR structures and comparison with X-ray.

20 March Dr. Gursky **Thermodynamic Methods I**

Protein Energetics - what is it good for?: Energetic-Structure-Function relationship in proteins. Thermodynamic versus mechanical description of macroscopic systems. Absolute temperature. Units and dimensions. Statistical weight and probability. Extensive and intensive variables. Energy as a state function. 1st law of thermodynamics. 2nd law, entropy S in spontaneous and equilibrium processes. 3rd law of thermodynamics.

27 March Dr. Gursky **Thermodynamic Methods II**

A Long Way to Gibbs-Helmholtz Equation: Energy E and enthalpy H. Heat capacity C_p and C_v: Microscopic meaning, typical values. Protein unfolding as a 1-st order phase transition. Gibbs free energy ΔG as a measure of protein stability. Typical values of ΔS, ΔH and ΔG for globular proteins. Entropy-enthalpy compensation in globular proteins.

03 April Dr. Gursky **Thermodynamic Methods III**

Cool and Hot Methods in Protein Thermodynamics: Gibbs-Helmholtz equation. Low-temperature protein unfolding as a test for Gibbs-

Helmholtz equation. Differential Scanning Calorimetry (DSC)– a direct method for thermodynamic analysis. Measuring protein heat capacity. Instrumental design. Calorimetric and Van't Hoff enthalpy, cooperativity. Application to single- and multidomain proteins and protein folding intermediates. Advantages and limitations of DSC.

10 April Dr. Gursky **Spectroscopic Methods I**

Spectroscopic Methods of Protein Thermodynamic Analysis: Gibbs distribution. Measuring protein stability ΔG by chemical unfolding. Van't Hoff plot; measuring enthalpy ΔHv by thermal unfolding. Indirect determination of the heat capacity increment ΔCp: heat unfolding at different pH, combination of thermal and chemical unfolding, low-temperature unfolding. Measuring small changes in stability of structurally similar proteins Le Chatelier's principle, applications to protein thermodynamics.

17 April Dr. Gursky **Spectroscopic Methods II**

Circular Dichroism (CD) Spectroscopic Analysis of Proteins: Review of light polarization. Definition of ellipticity. Review of light absorption; normal absorption, linear and circular dichroism. Physical origins of CD. Relation between CD and ellipticity. Typical values of protein ellipticity. Far-UV CD spectra of pure secondary structures. Thermodynamic analysis of a-helical proteins using CD. Spectral deconvolution and quantitative secondary structural analysis. Effects of protein tertiary structure on far-UV CD. Selection of the reference spectra.

24 April Dr. Gursky **Spectroscopic Methods III**

Infrared, Raman and Fluorescence Spectroscopy: Infrared spectroscopy: molecular vibration, stretching and bending modes. Principal IR bands for the peptide group as a function of secondary structure. IR polarization. Instrumental design of Fourier Transform IR spectrometers. Secondary structural analysis using FTIR. Differential FTIR. Raman spectroscopy: Stokes and anti-Stokes components of scattering. Intensity, resolution, applications of Raman to macromolecules and their complexes. Fluorescent spectroscopy: relation between absorption and emission spectra. Factors affecting fluorescent intensity. Steady state and kinetic measurements. Fluorescent quenching as a probe for solvent accessibility of protein chromophores. Probing folding states by fluorescent dyes. Fluorescent energy transfer.

Biophysics of Macromolecular Assemblies –GMS BY 776

Instructor: Dr. G.Graham Shipley

Introduction

- Course Outline
- Cell structure; nucleus endoplasmic reticulum, Golgi apparatus, mitochondria, lysosomes, membranes,etc.

- Protein structure/assemblies
- Lipids
- Membranes; membrane protein classes
- Protein-nucleic acid assemblies; chromatin, ribosomes, viruses
- "Double Helix;" BBC film of Crick, Watson, Wilkins, Franklin, etc. and structure of DNA

Protein Folding Motifs and Quaternary Assembly

- Introduction to general aspects of protein assembly
- Aims of this section of course
- Structural hierarchy, secondary structure, loops, motifs and domains
- Folding hierarchy, super-secondary motifs, αα, βαβ, ββ motifs
- Tertiary motifs
- Alpha structures, helix packing, helix dipole, helix-turn-helix motif, amphipathic helices, packing geometry, helix bundles and globin fold
- Beta structures, parallel vs. anti-parallel sheet, crossovers, loops, sheet topology diagrams, the Greek key
- Beta-alpha-beta structures, helical crossovers
- Folding domains, sequence characteristics, repeated sequence domains
- Domain vs. quaternary assembly
- Fold classification, the CATH database

Hemoglobin

- Myoglobin structure
- Hemoglobin quaternary assembly, symmetry of packing, cooperativity
- Hemoglobin/myoglobin sequence and secondary structure differences
- Subunit contacts and interactions
- Quaternary structure changes on oxygen binding
- Oxy vs. deoxy tertiary structure changes on oxygen binding
- Salt bridge interactions, role of penultimate Tyrosine, oxygen binding to iron
- Sequential tertiary structure changes, quaternary changes and cooperativity

Clathrin

- Overview of the endocytic pathway, receptors, fuzzy coat, coated pits and vesicles
- Structure of fuzzy coat, transition to pits and vesicle, symmetry of vesicle formation, 5 vs. 6 fold packing symmetry, Euler's theoremNegative stain EM of vesicle and cage structure
- Clathrin molecule, triskelion, domain organization
- Clathrin packing and assembly in vesicles
- Cryo-EM of cages and coats, location of clathrin domains, adaptor proteins, receptor interactions
- Clathrin molecular structure from x-ray crystallography, proximal leg and N-terminal domain

Spectrin

- Proteins of the erythrocyte membrane, cytoskeletal components

- Spectrin, Ankyrin, 4.1/4.2, Actin general features and interactions
- Assembly of Spectrin, repeated sequence domains, head-to-head vs. head-to-tail assembly
- Building the cytoskeleton, interactions
- Spectrin molecular details from x-ray structures, comparison with Actinin, structure of repeated unit
- Spectrin flexibility and conformational changes

Actin, Myosin and Tubulin

- Review of striated muscle, supramolecular structure, sliding filament model
- Symmetry of sarcomere and filaments
- Assembly of myosin, repeating motifs of tail region, coiled-coil structures • Myosin ATPase domain, molecular details from x-ray crystallography, domain structure, helix tail, ATP binding
- Assembly of Actin
- Molecular details from crystallography, domain structure
- Filament assembly, subunit interactions
- Actin-Myosin interactions
- Overview of cilia and organization of axoneme, protein of the axoneme
- Supramolecular structure of microtubules, helix geometry
- Tubulin dimmers, GTP/GDP binding
- Molecular structure, EM of tubulin sheets, subunit interfaces
- Molecular details, EM-crystallography, domain structure, nucleotide binding, subunit interfaces

Lipids Introduction

- Definitions and chemical classification of lipids
- Overview of lipid functions in cell and organisms
- Lipids – molecules with dual physical properties. HLB

(hydropholic lipophilic balance); water and hydrocarbon (oil) solubility

The Free Energy of Transfer from Water to Hydrocarbons • Lipid distribution (partition) between water and hydrocarbon

- The partition coefficient, $K_{w/o}$
- Data obtained from varying the number of $–CH_2-$ in a chain
- The free energy of transport from water to oil

$\Delta Gt_{w \rightarrow o} = RT \ln K_{w/o}$

- $\Delta G_{r\, w \rightarrow o}$ for methylene groups, $(-CH_2-)$ methyls

$(-CH_3)$, double bonds (-CH = CH-) and hydrophilic groups

Surface Behavior of Lipids

- Surface tensions and energy of cohesion
- Insoluble lipids – spreading and non spreading
- The spreading pressure
- Stable monolayers – Pockels – Langmuir balance
- Surface pressure/molecular area isotherms

- Unstable monolayers of soluble lipids
- Micelle formation and solubilization of hydrocarbons **Lecture 10: Structure of Lipids**
- Classification of lipids based on interaction with H_2O
- Structure and packing of aliphatic chains in lipids
- Phase transitions in lipids; aliphatic chain transitions
- Effects of polar substitution on phase transitions
- The non ideal liquid state of lipids; the concept of fluidity

The Mesomorphic State, a 4th State of Matter. Liquid Crystals

- Discontinuous changes in specific heat and volume between the solid crystalline state and the liquid state
- Definition of liquid crystals
- Structure of liquid crystals
- Classification of liquid crystals and ordered fluids
- Molecular motions, translations and translocation of lipids

The Lipids of Lipoproteins

- Introductions and historical prospective
- The classes of lipoproteins: chylomicrons (CM), VLDL, LDL, HDL, albumin • The lipids of lipoproteins – classification and physical properties –phosphatidyl choline (PC), triacylglycerol (TG), cholesterol (C), cholesterol esters (CE)
- The interaction of lipids in lipoproteins

Phase Behavior of the Lipoprotein Lipids

- The phase rule
- The PC- H_2O system
- The PC-C-H_2O, PC-CE-H_2O, PC-TG-H_2O system
- The lipoprotein lipid phase diagram
- Location of phases within a lipoprotein

The Apoproteins

- The major apoproteins AI, AII, AIV, B, C, CI, CII, CIII and E
- Exchangeable and non exchangeable apoproteins
- Secondary and tertiary structure of apolipoproteins, amphipathic alpha helices (AAH) and amphipathic B strands (ABS)
- Interaction of apoproteins with lipids

Lipoprotein Assembly, Plasma Conversions and Uptake – Physical Considerations

- Synthesis and secretion of apoB containing lipoproteins CM, VLDL
- Plasma conversions of VLDL to LDL – LDL uptake
- Formation of HDL

Biophysics of Macromolecular Assemblies –GMS BY 777

Instructor: Dr. G.Graham Shipley

Cell Membranes and Membrane Proteins

- Review of overall membrane organization
- Cell membranes; plasma membrane, organelle membranes
- Membrane functions
- Membrane structure; average structure, localized structural domains
- Membrane composition; lipids, proteins
- Membrane lipids; organization, bilayers, distribution, dynamics • Membrane proteins; organization, dynamics, functional classes • Structural motifs of membrane proteins?

Bacteriorhodopsin

- Halobacteria, H. Halobium, energetics, purple membrane
- Bacteriorhodopsin, light activated proton pump
- Early studies; isolation, chemical characterization
- Electron microscopy, x-ray diffraction, hexagonal arrangement
- Electron crystallography; Henderson/Unwin, 2D and 3D, transmembrane alpha-helical bundles • Labeling, neutron diffraction
- Retinal location, orientation
- Helix connectivity
- High resolution studies; electron and x-ray crystallography
- Proton channel, photocycle and pump mechanism

Photosynthetic Reaction Center

- Plants, bacteria, energy transduction, photosynthesis
- Structure of R. viridis reaction center; L, M, H and cytochrome subunits
- Arrangement of prosthetic groups; heme, bacteriochlorophyll, bacteriopheophytin, carotenoid, quinine
- Electron flow
- Structure of bacterial light-harvesting complex; protein and pigment organization
- Structure of mitochondrial cytochrome bc1 complex; protein subunit structure

Porins

- Porins in Gram-negative bacterial and mitochondria
- E. coli porins; PhoE, OmpF and OmpC
- Early structural studies; electron microscopy of OmpF and PhoE, trimers
- Transmembrane beta-barrel structures
- X-ray structures of OmpF, PhoE an Dr. capsulatus porins; 16-strand barrels, channel structure
- X-ray structure of OmpA; 8-strand barrel
- X-ray structure of FepA, 22-strand barrel, and Ompla (12 strand barrel
- Structure of maltoporins; 18-strand barrel, role of loops, structure of pore, sugar transport

Bacterial Toxins

- S. aureus alpha hemolysin; cell lysis, oligomerization
- Structure of alpha hemolysin; monomer structure, heptamer structure
- Pore structure; 14-strand beta-barrel

- Anthrax toxin; subunit structure, mechanism of action
- Structure of protective antigen; domain structure of monomer, heptameric assembly, 14-strand beta-barrel?
- Structure of cholera toxin

Influenza Virus Hemagglutinin

- Influenza virus; structure and mode of action
- Influenza virus hemagglutinin; receptor binding and membrane fusion activities • Influenza virus hemagglutinin structure; bromelain treatment, trimeric assembly, coiled-coil domain, HA1 and HA2, receptor binding site, glycosylation sites, fusion activation site
- Influenza epidemics and pandemics; antigenic drift and shift, relation to structure • Low pH structure of influenza virus hemagglutinin; conformational changes, fusion model

Potassium Channel

- Membrane ion channels, Na^{+}, K^{+}, Ca^{2+}
- Potassium channels; voltage-gated and ligand-gated
- Structure of KcsA K^{+} channel pore; selectivity filter, ion conduction
- Structure of calcium-gated MthK K^{+} channel; gating model
- Structure of chloride channels

Protein-Nucleic Acid Interactions

Chromatin

- Procaryotic and eukaryotic chromatin
- DNA structure; review double helix structure, higher order folding
- Eucaryotic chromatin; DNA/histone complexes
- Histones; sequence, structure, evolution, assembly
- Nucleosomes; “beads on a string,” nucleosome core particle, histone octamer
- Nucleosome/core particle structure; electron microscopy, x-ray diffraction
- Nucleosome core particle; x-ray crystallography (Klug/Richmond), DNA-histone interactions, DNA superhelix, histone octamer

Ribosomes

- Ribosomes and protein synthesis
- Procaryotic and eukaryotic ribosomes; subunits, ribosomal RNS and ribosomal proteins, disassembly/reassembly (Nomura)
- E coli ribosome structure; cross lining, electron microscopy, immunoelectron microscopy
- E coli 30S and 50S subunits; neutron scattering, distance measurements, triangulation
- E coli ribosome structure; cryoelectron microscopy, 3D crystals
- High resolution x-ray structures of 50S and 30S subunits; structure of complete ribosome
- Mechanism of protein synthesis

Viruses

- Introduction; examples of DNA/RNA ss/ds viruses, overall structure, nucleic acid, protein capsid, membrane envelope
- Virus shape; spherical, rod-shaped, complex
- Virus structure; core/shell model, symmetric protein shells (Crick/Watson)

- Tobacco mosaic virus; rod-shaped RNA virus, electron microscopy, helical structure, RNA and protein helices
- TMV protein; disks, cylinders, helix, TMV protein disk structure
- TMV structure; x-ray fiber diffraction, RNA helix, disk-helix transition, RNA-protein interactions
- TMV assembly; initiation sequence, disk-helix transition

Spherical Viruses

- Spherical viruses; cubic symmetry, polyhedra, icosahedral symmetry
- Examples; adenovirus, herpes virus, polyoma virus, etc.
- Icosahedral symmetry; simple (T=1) and complex (T-3) icosahedra, Caspar/Klug quasi-equivalence
- Satellite tobacco necrosis virus (STNV); T=1 capsid structure, symmetry
- Tomato bushy stunt virus (TBSV); T=3 capsid structure, symmetry, quasiequivalence
-

Module V: Physiology of Specialized Cells GMS FC707

Course Director:
Christopher Gabel, Ph.D., Physiology and Biophysics

Course description:
This course is one of the elective course modules (Module V) of the Foundations in Biomedical Sciences Curriculum. Knowledge of cellular and molecular physiology is critical to understanding the higher order functioning of tissues, organs and organ systems. The objective of Physiology of Specialized Cells is to discuss the specialized adaptations of selected cells that help them to function in their respective tissues and organs. This course will also provide a frame work to bridge the gap between the biochemistry and the molecular and cellular biology that students have acquired in the core modules and organ physiology and pharmacology that will be addressed in the second year. Physical and chemical principles will be presented in the context of physiological concepts and further explained with clinical examples. The course will cover basic cellular processes including: Diffusion, Homeostasis, Ion Channels and Excitable Membranes, and Solute Transport. The course will highlight the specific adaptations of various cell types that allow them to perform their distinct physiological functions.

Faculty:
Dr. David Atkinson atkinson@bu.edu Physiology and Biophysics
Dr. Fernando Garcia-Diaz fgarcia@bu.edu Physiology and Biophysics
Dr. Scott Downing sdowning@bu.edu Pharmacology
Dr. Clint Makino cmakino@bu.edu Physiology and Biophysics
Dr. Michael Rynkiewicz rynkiemj@bu.edu Physiology and Biophysics
Dr. Chris Gabel cvgabel@bu.edu Physiology and Biophysics

Lecture #1: Introduction to Cellular Homeostasis and Diffusion: Microscopic Theory
Dr. Gabel

Lecture #2: Solute Transport, Water Movement and Osmotic Pressure
Dr. Garcia-Diaz

Lecture #3: Electrical Consequences of Ionic Gradients
Dr. Downing

Lecture #4: Specialized Cell: Hepatocyte cellular transport
Dr. Atkinson

Lecture #5: Generation and Propagation of Action Potentials
Dr. Downing

Lecture #6: Ion Channels and Ion Channel Diversity
Dr. Atkinson

Lecture #7: Specialized Cell: Auditory system (Hair cells)
Dr. Gabel

Lecture #8: Specialized Cell: Visual System (photoreceptor cells)
Dr. Makino

Lecture #9: Specialized Cell: Muscle cell function and structure
Dr. Rynkiewicz

Lecture #10/11: Student Presentations
Dr. Gabel