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

Fall Semester, 2022:

Location: W310 (Center for Advanced Biomedical Research, CABR, 700 Albany St.)

Course format: The course will be conducted through both a didactic and a tutorial mechanism. For tutorial-based classes, 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. Preparation and participation in the course discussions will also contribute to grades.

Schedule:

19 September	Dr. Atkinson	**Introduction,** Perspectives and background of structural biology. Symmetry in structural biology. **Symmetry Operations: Point and Space Groups,** Rotation and mirror symmetry, translation, glide, and screw operations
26 September	Dr. Atkinson	**Fourier Theory in Structural Biology** Waves, Fourier series. Fourier and inverse Fourier Transforms. **Fourier Theory in Diffraction:** Importance in Structural methods, Convolution, Correlation
3 October	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.
10 October	Indigenous Peoples Day (class to be held on Tuesday 11th)	
11 October	Dr. Atkinson	**X-ray Crystallography II** **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.
17 October	Dr. Rynkiewicz	**X-ray Crystallography III** **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.
24 October	Dr. Rynkiewicz	**X-ray Crystallography IV** **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.
31 October	Dr. Rynkiewicz	**X-ray Crystallography V** **The Phase Problem:** Phase determination: Multiple isomorphous replacement, multiple anomalous dispersion, molecular replacement, direct methods. Phase improvement: Solvent flattening, histogram matching, non-crystallographic averaging.
07 November	Dr. Rynkiewicz	**X-ray Crystallography VI** **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.
14 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
21 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 EM hands-on intro to imaging.
23 - 26 November		Thanksgiving Break
28 November	Dr. Bullitt	**Structural Electron Microscopy III** **3-Dimensional Image Reconstruction, Overview and Single Particle Analysis (SPA):** The projection theorem and its application to 3D structural analysis of electron micrographs Cross correlation and Bayesian alignment Common lines methods in reciprocal or real space Single Particle Analysis Classification methods
05 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
12 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 2D crystals analyzed from electron diffraction and/or images