Hydrostatic pressure is a force that all organisms experience. Biomolecules and water together seek to minimize total system volume. Under intense pressure, this leads to interesting and non-intuitive results since biomolecules are never perfectly packed at the atomic level. Rather than simply compressing, molecules with functionally important voids and cavities tend to change conformation, dissociate, and even unfold to minimize volume. Lipid mesophases also seek to minimize volume, and it is thought that functionally important lipid configurations such as those required for cell division are more pressure sensitive than standard lamellar structures. Liquid-liquid phase separation too is pressure sensitive in ways that are not yet fully understood. The characterization of structure under intense pressure has consequently gained considerable recent interest. Beyond being a tool for fundamental biophysics, high-pressure structure provides important insights to both the extremophile (deep life) and high-pressure food processing communities.

Though technically challenging from an engineering standpoint, high-pressure BioSAXS (HP-SAXS) is now routinely available to non-specialists and can be performed in high volume at the CHEXS HP-Bio facility. HP-SAXS is available in two modes: the “batch” method, which can achieve up to 400 MPa (4000 bar) and is most appropriate for titrations and generation of phase diagrams, and chromatography-coupled SAXS (HP-SEC-SAXS), which is better suited for mixtures and aggregation-prone samples at pressures up to 100 MPa (1000 bar). Both techniques can be conducted at temperatures < 4° C to 60° C (HP-SEC-SAXS) and 80° C (batch HP-SAXS) respectively, spanning typical ocean floor to hydrothermal vent conditions. Batch HP-SAXS at CHEXS is also possible under strictly anaerobic conditions and has been used in the presence of dissolved gasses. This lecture will cover the basics of high-pressure BioSAXS, giving recent examples and recommended protocols. Prospects for future developments such as oscillating batch samples, temperatures above 100° C and below 0° C and reaching beyond 100 MPa for HP-SEC-SAXS will also be discussed.