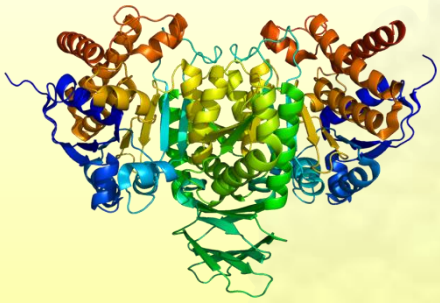
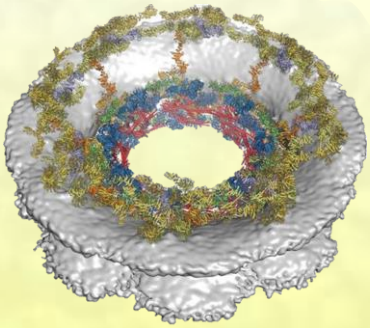
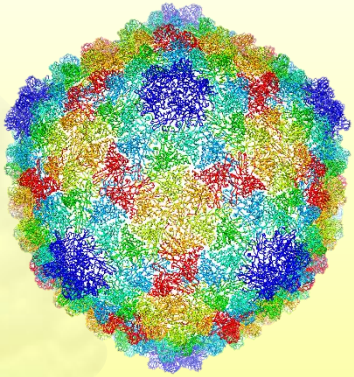
Biological Imaging - X-rays and Electrons

@ SSRL – SLAC







Biological imaging – using x-rays, electrons or a combination – is available to the scientific community through an integrated facility at SLAC National Accelerator Laboratory. The SSRL Structural Molecular Biology program provides four primary synchrotron x-ray capabilities: macromolecular crystallography (MC), small angle x-ray scattering/diffraction (SAXS), x-ray spectroscopy (XAS/XES), and x-ray microXAS imaging, with SSRL's high x-ray brightness beams. The SLAC-Stanford Cryo-EM Facilities provides state-of-the-art instrumentation, computational capabilities and support facilities for cryo-EM and cryotomography studies. Along with vigorous R&D in enhanced data collection, data management and data analysis, both activities support remote access, and user-friendly, real-time and on-line data interpretation. The innovations are closely coupled to projects aimed at solving forefront science problems. The synergy between the x-ray and cryo-EM facilities enables tackling challenging systems relevant to the DOE-BER and NIH missions. These programs seek to enhance the user community through excellent support, training and dissemination.

Supported by the DOE Office of Biology and Environmental Research, the NIH National Institute of General Medical Sciences, and non-Federal funding

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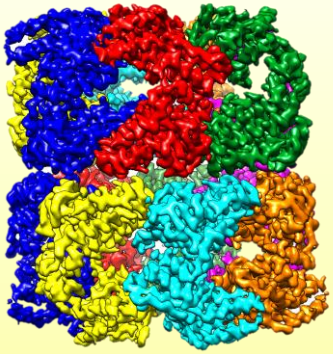
SMB / Cryo-EM User Administration

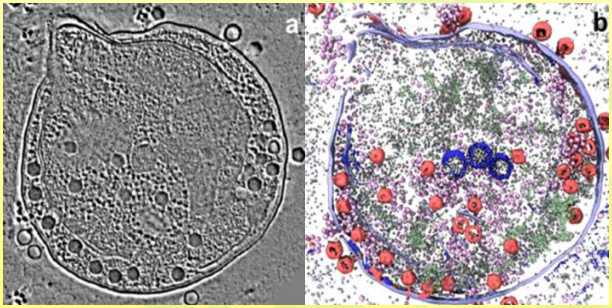
Cryo Electron Microscopy

imaging structure at the near-atomic level

Cryo Electron Microscopy (Cryo-EM) uniquely captures individual instances of structures of biological assemblies, in their natural heterogeneity, as they occur in solution. Cryo-EM can average sets of macromolecules whose structures are the same, to derive snapshots of conformational dynamics.



GroEL is a 14-mer chaperonin required for proper folding of many proteins. Cryo-EM has shown that very particle has a unique configuration of (at least) three different GroEL conformations among its 14 subunits.



Cryo electron tomography (Cryo-ET) using phase plate optics can reveal the 3D structure of a vitrified whole cell volume, without staining or chemical fixation.

A tomogram slice of phage-infected cyanobacterium (left) and its feature annotation (right) showing infecting phages outside, and a variety of newly assembled phage intermediates inside the cell (pink), subcellular structures like ribosomes (magenta), carboxysomes (blue), light harvesting machinery, etc.

Data from Cryo-EM, SAXS and MC data provide highly complementary structural information at different resolution; the seamless integration of these data and information provide a foundational platform for studies of biological systems.

The SLAC-Stanford Cryo-EM Facilities includes 3 high-end Titan Krios and 1 Talos Arctica cryo-EM instruments, computing infrastructure, laboratories for grid and cryo-sample preparation, and tissue culture laboratories with biohazards capabilities. An Aquilos cryo-FIB instrument will be available in 2020.

https://cryoem.slac.stanford.edu/

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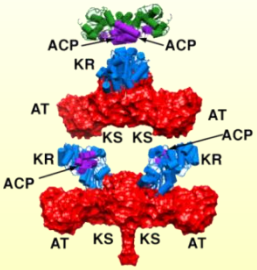
Small Angle X-ray Scattering

solution conformation of macromolecules



Polyketide Synthase

Small angle x-ray scattering (SAXS) from solutions of macromolecules, their complexes and assemblies provides 3D structural information at nm resolution, enabling interpretation of atomic-resolution structures and complex arrangements in a physiological context, informing molecular interactions, domain folding properties and structural flexibilities **in solution**.

SSRL's SAXS BL4-2 provides instrumentation for automated high-throughput chromatography-coupled solution x-ray scattering, **time-resolved** scattering in the msec time regime, and instrumentation for diffraction studies of **partially ordered** systems, *e.g.* lipids, fiber and membranes.

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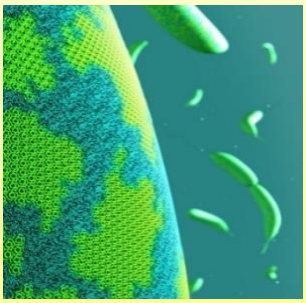
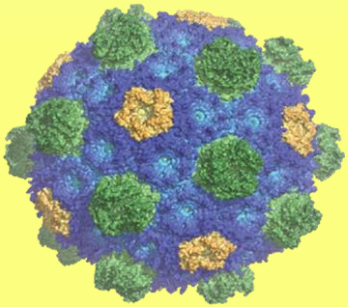


Protein shell surface layer of Caulobacter crescentus; amorphous and crystalline forms

Macromolecular Crystallography

3D structural information at the atomic level



Bacterial microcompartment shell of proteins surrounding an enzyme core; from MC and cryoEM

SSRL's four MC stations are fully **remote access** (run the experiment from your home institution) fully automated, using robotics for crystal mounting and screening. Diffraction images are **automatically analyzed**, indexed and scored. BL12-2 (micro-focus) and BL9-2 have high-speed Pilatus detectors for fast shutterless data collection and low-dose crystal searches. A next-generation **microfocus station**, BL12-1 will be available in 2020. A **broad bandpass** capability providing exceptionally bright microbeams and a high-speed Eiger16M detector (**133 Hz**) will support crystal injectors and other emerging techniques.

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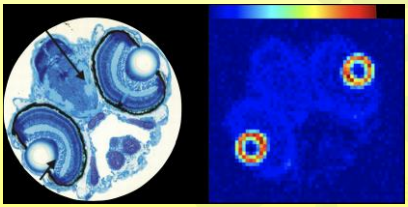
http://www-ssrl.slac.stanford.edu/smb/index.html

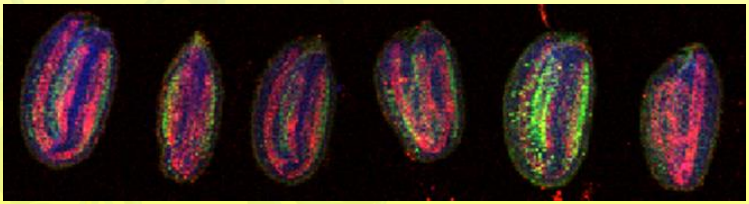
X-ray Micro-XAS Imaging

electronic & geometric structure of metal sites

The high brightness x-ray beams at SSRL's beam lines enable x-ray fluorescence-based imaging techniques, over a wide range of length scales, for research projects in fields including biological, environmental, and bio-geological science. Focusing approaches provide flexible beam sizes, ranging from sub-micron to a few hundred microns. These techniques, used at beam lines 2-3, 6-2, 10-2 and 14-3, reveal **where** different chemical elements are located, **what** elements are present in the sample, the relative **amount** of each element, and the **chemistry** of the element at specific locations. They can be applied on a wide variety of samples, *e.g.* from soils, plants, and tissues to cells. To obtain 3-dimensional images, x-ray fluorescence tomography (μm to mm spatial resolution) is also frequently used.



Quantitative Hg distribution in the outer layer of the eye lens of MeHg-treated zebrafish



X-ray microXAS imaging screening of Arabidopsis sp. seeds for variation in metal phenotypes with genetic variation [Fe (red), Mn (green), Zn (blue)

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https://www-ssrl.slac.stanford.edu/smb/index.html

Robust training programs for the novice and experienced user community, focus on the experimental and theoretical aspects of data measurement, analysis and chemical/structural biological understanding, for all techniques and science areas.