A Complete Micro-Electron Diffraction (MicroED) Solution for Fast Structure Determination of Macromolecules and Small Molecules

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X-ray diffraction using synchrotron radiation established a routine single-crystal structure determination workflow for small and macromolecules; however, these experiments require large, well-ordered crystals (50-100 μm). Growing large protein crystals is a critical bottleneck that is either time-consuming or challenging to overcome, and often smaller crystals are more attainable. Although microfocus beamlines can analyze crystals as small as 10-50 μm, they are prone to radiation damage or diffract X-rays weakly, limiting achievable resolution. Electrons are more advantageous than X-rays for the analysis of very small crystals (well below 1 μm in size) because accelerated electrons scatter more readily than X-rays, resulting in a stronger signal from thinner samples. Micro- or nano-crystal electron diffraction (MicroED) is thus well-suited for the analysis of small crystals, producing high-resolution 3D structures of small chemical compounds or biological macromolecules.

MicroED experiments do not require a specialized synchrotron facility; instead, cryo-TEM equipment that fits in a standard laboratory can be used. MicroED experiments are carried out in a vacuum, which reduces background scatter. The use of cryogenic temperatures protects the sensitive crystal sample from radiation damage, which would otherwise result in rapid degradation. Because only nanoscale crystals must be grown, this method significantly shortens the sample preparation process and decreases the amount of pure product required. Furthermore, intermediate-sized crystals that are too large for MicroED but too small for X-ray crystallography can be solved by physically breaking up the crystals or by thinning with a cryo-focused ion beam instrument (cryo-FIB).

In this session, we will talk about our complete MicroED workflow, focusing on innovations in Cryo-TEM hardware, sample screening, and data collection. You will learn how MicroED allows for the rapid determination of the 3D structure of small chemical compounds and biological macromolecules using a cryo-TEM instrument outfitted with a specially designed hardware/software package to efficiently collect diffraction datasets of nanocrystals. We will highlight innovations and advances in hardware components including optics, stage mechanics, and detectors as well as software for automated and higher-throughput data collection. Together, we will show how innovations in the field of MicroED allow for data collection that can be completed in a matter of minutes for high-resolution structure determination in a variety of fields, including structural biology, medicinal chemistry, and other organic and inorganic chemistry disciplines with nanocrystalline samples.