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Electron diffraction of 3D nanocrystals: an emerging technique in structural biology

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Electron diffraction of 3D nanometre sized crystals, most commonly named microED, has recently emerged as a new technique to solve the structure of both small organic molecules and proteins (1–3). MicroED is clearly a promising technique in structural biology, both quite easy to implement and very complementary to X-ray crystallography and single particle cryo-EM. Electrons are actually a very interesting probe for small samples as they strongly interact with matter, and more importantly, they deposit much less energy than X-rays per diffracted particle (4). The drawback of strong interaction is the presence of multiple diffraction events that make the relationship between diffracted intensities and structure factor much complicated than with the kinematical theory of diffraction. Nevertheless, sub-atomic resolution data can routinely be collected for small organic compounds and data up to 1.5 Å resolution can be obtained from protein crystals. Phasing can be done by either direct methods for sub-atomic resolution data or by molecular replacement and refinement can easily be performed with programs developed for X-ray crystallography, such as Refmac or Shelxl. When the number of atoms is small, multiple diffraction can be taken into account at the refinement stage to improve the final Coulomb potential map (5, 6). During the past few years, microED has been very successful for solving the the structures of small organic molecules, metallo organic framework and proteins up to 100-200 kDa. However, a quick look at the Protein Data Bank shows that among the 66 protein structures determined so far by microED, only 10 are not model proteins, and just 2 were solved with models sharing less than 99% sequence homology. 9 years after the first articles marking the emergence of microED in structural biology (7, 8), this indicate that the technique is still far from routinely used. Based on examples either studied at IBS, thanks to our hybrid pixel direct detector mounted on an F20 200 kV electron microscope, or picked up in the literature, I will give an overview of the main results of the technique but also focus on remaining bottlenecks.

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