Beyond MicroED: *Ab Initio* Structure Elucidation using 4D-STEM

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Microcrystal electron diffraction (microED) has recently morphed into an increasingly mainstream technique in structural chemistry [1]. Its ability to interrogate nanocrystals orders of magnitude too small for conventional X-ray diffraction has enabled solid-state structure elucidation of several species previously considered impossible to solve using X-ray crystallography [2, 3]. Nevertheless, selected area aperture-enabled microED remains thwarted by the presence of disordered, overlapping, or otherwise poorly diffracting domains, all of which routinely conspire to diminish data quality. Just as insufficient crystal size historically stymied conventional X-ray methods, these nanoscale defects frequently prohibit structure solution using classical microED.

To overcome this, we apply 4D scanning electron microscopy (4D-STEM) [4] in conjunction with electron diffraction tomography [5] to interrogate crystal structures spanning a wide gamut of chemical space, including beam-sensitive organometallic complexes and biomolecular organic compounds. Our results represent the first 4D-STEM structures phased *ab initio* by direct methods. Unlike standard microED, data acquisition in 4D-STEM is not constrained by the shape or size of the SA aperture. Instead, 4D-STEM enables the *ex post facto* construction of bespoke virtual apertures [6], allowing for precise real-space localization of exactly which domains of crystal contributed to productive Bragg diffraction in reciprocal space (Figure 1). We refer to these regions as coherently diffracting zones (CDZs). This empowers us to discard unwanted signal from poorly diffracting domains, rotationally misoriented CDZs, or coincident lattices, effectively enabling the collection of viable datasets from specimens unusable for standard microED (Figure 2). Several strategies to parse these data—such as *k*-means clustering, center-of-mass analysis, and directly indexing sparse diffraction patterns using algorithms originally designed for XFEL experiments [7]—will be discussed.

Our analysis unveils that CDZs in molecular crystals form an intricate topography of unpredictably distributed striations. CDZs appear as randomly shaped slivers often embedded within relatively Bragg-silent domains, even in nanocrystals anticipated to contain well-ordered, monolithic lattices. The ubiquity of these imperfections indicates sharp deviation from the defect-free crystals assumed in computational simulations of electron diffraction patterns [8], providing an explanation for the current chasm between theory and experiment [9]. We will also discuss the beam-induced evolution of CDZ contours as a consequence of propagating radiolytic damage. [10]

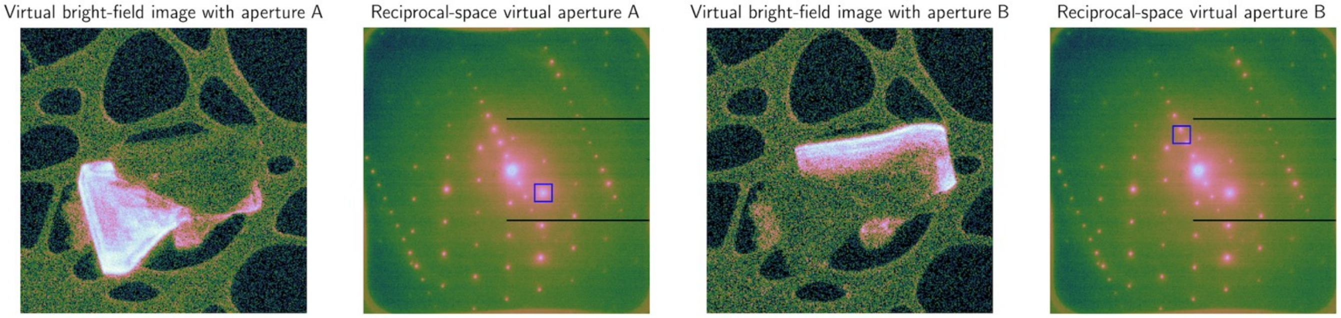

Figure 1. Virtual bright-field 4D-STEM images generated by placing virtual apertures (dark blue squares) over specific Bragg peaks in reciprocal space, demonstrating the isolation of independent coherently diffracting zones (CDZs).

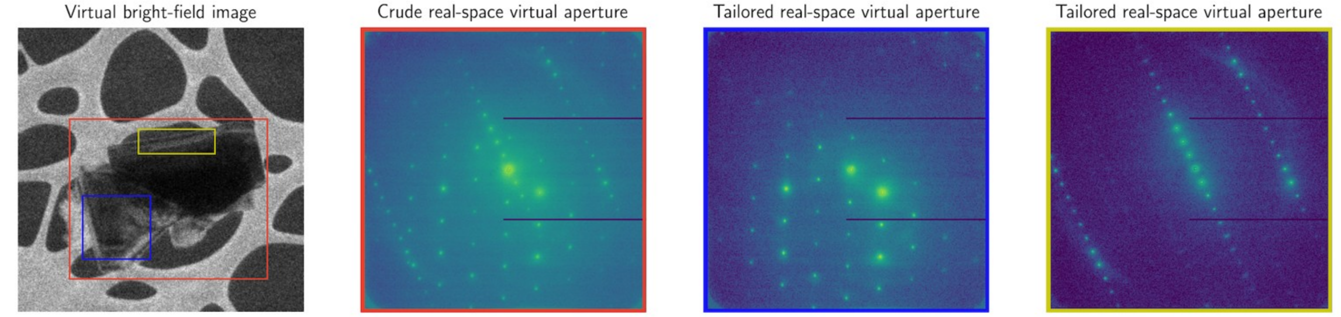

Figure 2. Virtual bright-field 4D-STEM image with three virtual apertures (red, yellow, and blue boxes) superimposed on the crystal of interest. Each virtual aperture generates a distinct diffraction pattern. The large red box—roughly equivalent to a crude SA aperture—yields an aggregate diffraction pattern consisting of two coincident lattices, indicating a polycrystalline sample which would be swiftly discarded as defective if using standard microED. Nevertheless, the smaller blue and yellow boxes demonstrate a quick and powerful deconvolution of these lattices simply by using tailored real-space virtual apertures.

References

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