Applying the Semi-Automated MicroED Processing Pipeline, AutoMicroED

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Although still a relatively new cryogenic electron microscopy (CryoEM) technique, microcrystal electron diffraction (MicroED) is rapidly growing in popularity [1]. MicroED is analogous to X-ray crystallography in that it is used to determine atomic structure from three-dimensional crystals, but it utilizes a transmission electron microscope as the base platform. Add-on modules for MicroED data collection are available for most new microscopes and many older generation instruments can also be modified to collect data. In addition, standalone electron diffractometers have recently become available. Due to the widespread availability of instruments that can be used for MicroED collection, MicroED has been used in a broad variety of fields such as natural products research and drug discovery [2-3] and has thus far been used to determine structures of proteins, small molecules, and peptides [4-6].

Despite the growth of the MicroED field, the processing methods for MicroED data are still cumbersome. Several software packages exist but largely require manual or semi-manual curation and familiarity of the user with X-ray crystallography software packages. This creates a steep learning curve for those new to the field and slows the time for structure elucidation. Recently, we presented our solution to this problem in the form of a semi-automated MicroED data processing pipeline called AutoMicroED [7]. AutoMicroED allows users to quickly, and easily, go from data to structure using a single command line command prompt even for heterogenous or mixed sample datasets (Figure 1). The pipeline runs through image conversion, indexing, integration scaling, merging and final structure determination, and is functional for both small and macromolecules. AutoMicroED is a useful processing tool for those new to the field of MicroED and also permits experienced users to modify or optimize any standard parameters. Our previous results demonstrated the use of AutoMicroED with molecules of known structure. Here, we present the application of AutoMicroED used with a molecule of previously unknown structure [8].

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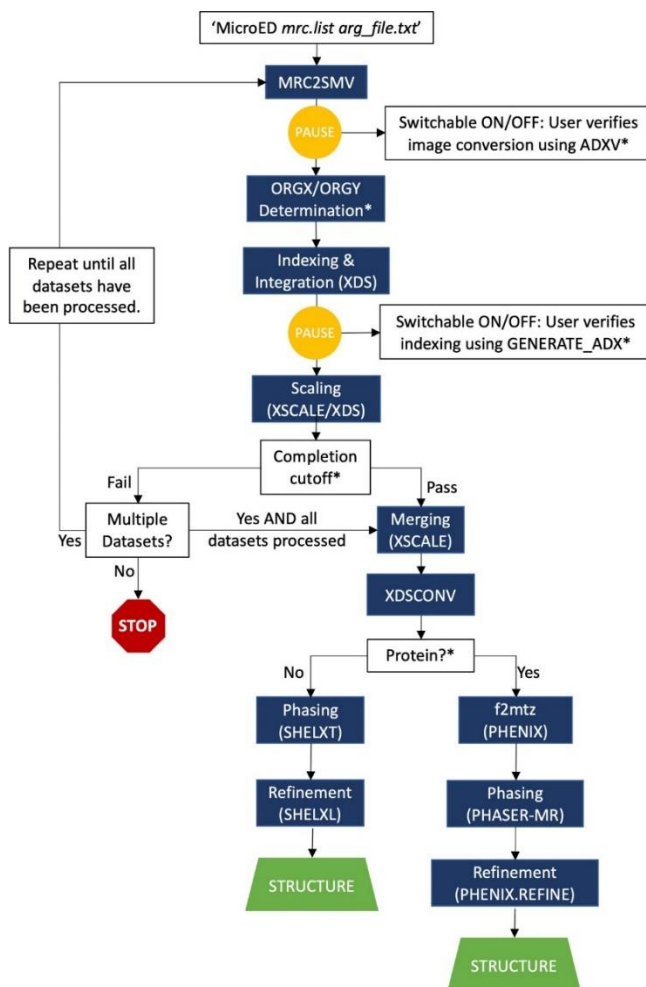
Figure 1. AutoMicroED data processing workflow showing full pipeline and decision points assuming MRC file input. The * indicates points that the user can control interactive or noninteractive status using the arg_file. Bypassing these control points allows direct automated structure determination following a single command prompt. Adapted from [7].

References:

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