

Micro-crystallisation of Photosynthetic Reaction Center for Time-Resolved Serial Femtosecond Crystallography at an X-ray Free Electron Laser

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Advances at X-ray free electron lasers (XFELs) are broadening the scope of this technology and allowing experiments with more varied and difficult protein targets. One remaining bottleneck however is the production of micro-crystals suitable for delivery to the XFEL beam. Through a series of experiments at the Linac Coherent Light Source, California, we have optimised two different methods for micro-crystal production of the photosynthetic reaction center from *Blastochloris viridis*. One, in which we crushed macro-crystals mechanically through vortexing, and another in which we grew homogeneous samples of micro-crystals using a micro-seeding method. These methods have led to an increased crystal hit-rate and thus decreased sample usage compared to previous experiments on the same protein. Our micro-crystals diffracted to 2.85 Å, and were suitable for a time-resolved experiment, in which we visualised the protein at two time-points of the photosynthetic reaction. The micro-crystallisation techniques described will be valuable additions to the small but growing library of techniques available for structural biologists performing serial crystallography experiments at an XFEL.