MS01-P10 | Liquid Application Method for time-resolved Analyses (LAMA) by serial synchrotron crystallography

Mehrabi, Pedram (Max-Planck-Institute for Structure and Dynamics of Matter, Hamburg, GER); Schulz, Eike (Max Planck Institute for Structure and Dynamics of Matter (MPSD), Hamburg, GER); Agthe, Michael (Universität Hamburg, Hamburg, GER); Horrell, Sam (University of Hamburg, Hamburg, GER); Bourenkov, Gleb (EMBL Hamburg, Hamburg, GER); von Stetten, David (EMBL Hamburg, Hamburg, GER); Leimkohl, Jan-Philipp (Max Planck Institute for Structure and Dynamics of Matter (MPSD), Hamburg, GER); Schikora, Hendrick (Max Planck Institute for Structure and Dynamics of Matter (MPSD), Hamburg, GER); Schneider, Thomas (EMBL Hamburg, Hamburg, GER); Pearson, Arwen R. (University of Hamburg, Hamburg, GER); Tellkamp, Friedjof (Max Planck Institute for Structure and Dynamics of Matter (MPSD), Hamburg, GER); Miller, R. J. Dwayne (Max Planck Institute for the Structure and Dynamics of Matter, Hamburg, GER)

With the advent of serial crystallography and new light sources time-resolved crystallography has recently seen a resurgence. Since the majority of enzymatic reactions occur in the range hundreds of milliseconds, synchrotron light sources are a clear alternative to XFELs for these systems due to their wide distribution and their simpler experimental setup.

Previously we combined an established fixed target mount (chip), with a novel "*Hit And REturn"* (HARE) approach for data acquisition. Due to the HARE algorithm time delays from a few milliseconds to several minutes can be acquired with no increase in data collection time [1].

Extending the HARE approach with the use of a piezo droplet injector circumvents the use of physical or chemical triggers for reaction initiation, such as temperature-, pH jumps, or light pulses for systems that are not naturally amenable to light activation. Our "*Liquid Application Method for time-resolved Analysis"* (LAMA) in combination with the HARE approach uses minimal amounts of protein (1-3 mg) and ligand (1.5-3 µl) per time point, making this scheme tremendously material efficient. Using Glucose Isomerase as a model system we were able to observe an open ring conformation of a glucose molecule in a reaction that spans several minutes in the enzyme. These newly developed methods are highly flexible, can be easily adapted to synchrotron and XFEL beamlines and are amenable to the majority of enzymatic systems, which now become accessible to time-resolved studies.

[1] Schulz, E. C., Mehrabi, P., Müller-Werkmeister, H.M. *et al Nat. Meth.* - 2018 doi:10.1038/s41592-018-0180-