Synchrotron X-ray scattering from biomacromolecular solutions –new developments and results

Dmitry I Svergun

European Molecular Biology Laboratory (EMBL), Hamburg, Germany

svergun@embl-hamburg.de

Small-angle X-ray scattering (SAXS) is an established method to study the overall structure of biological macromolecules and complexes in solution. The last decade saw a major progress in SAXS data analysis methods and instrumentation. Large scale structural studies in molecular biology are now possible on high brilliance synchrotrons thanks to the automation of the experiment, data processing and interpretation. SAXS is directly applicable to medical formulations raising the importance of the technique for pharmaceutical industry. SAXS is often combined with other structural methods like crystallography, NMR and electron microscopy, and also with computational, biophysical and biochemical techniques to build hybrid models. In classical applications, SAXS generally yields low resolution quaternary structure but, very importantly, the method can also help to analyze equilibrium mixtures and to visualize flexible portions of the structures, not seen by the high resolution methods.

Here, recent developments at the high-brilliance synchrotron P12 beamline of the EMBL (PETRA III storage ring, DESY, Hamburg) will be presented. P12 has been designed and optimized for scattering experiments on macromolecular solutions. The sample environment can be rapidly exchanged from the standard robotic sample changer to conduct other types of SAXS experiments such as on-line purification with size exclusion chromatography, scanning SAXS, microfluidic chips etc. The photon-counting PILATUS 6M detector enables a background-free detection of weak scattering signals. Versatile and flexible sample environment system coupled with automated pipeline for data acquisition and analysis provides near real-time assessments of overall parameters and the generation of low-resolution models within minutes of data collection.

A recently commissioned multilayer monochromator delivers the flux of 5×10^{14} photons per second on the sample. Together with the new Dectris EIGER 4M detector that operates at 750 Hz, reliable data from biological solutions can be routinely collected as a millisecond frame rate.

The beamline is also equipped with a Biokine SFM-400 Stopped Flow apparatus for kinetic studies. A mixing dead time of only three milliseconds was achieved to monitor (un)folding and (dis)assembly reactions. Additionally, a continuous flow microchip and laser triggering devices are being developed to further reduce the dead time and allow sub-millisecond time resolved SAXS experiments. Pilot time-resolved experiments conducted at P12 will be presented.