Microsymposium

MS77.003

Optimizing buffer conditions to improve crystallizability and SAXS measurement

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The Sample Preparation and Characterization (SPC) facility at EMBL Hamburg is situated next to the PETRA3 beamlines for protein crystallography and small angle scattering (SAXS) that are operated by EMBL. The facility is equipped with molecular biology and biophysical instrumentation to carry out purification and characterization of macromolecular samples. It serves a mixed community of local EMBL scientists, beamline visitors and scientists from the European Union research area. Most incoming samples are destined for high-throughput crystallization at the facility or characterization by SAXS. The facility offers standardized quality control reports on each incoming sample, including characterization by mass spectrometry and Thermofluor. Based on this report, local staff can suggest and perform optimization protocols that increase the stability of the sample. For instance, Thermofluor screens(1) were developed that probe the effect of buffers and additives that are commonly used in sample preparation. These systematic screens provide a high-throughput method to identify stabilizing conditions for sample purification, storage and structural characterization. This technique has been also a valuable asset providing a high-throughput method for assessing the crystallizability of proteins by screening for conditions which contribute to the protein sample homogeneity, stability and solubility. The home-made screens have been tested on more than 200 different protein constructs at SPC facility. The aim of the SPC facility is to integrate off-line biophysical techniques with synchrotron beamlines, to offer the European user community a full package for sample characterization. The facility is especially geared towards cell biologists with little experience in structure determination. Expert staff is available to help to plan, perform and interpret biophysical experiments. It is possible for users to book SPC equipment together with their synchrotron beamtime, for protein purification, circular dichroism and isothermal calorimetry. Funded access to the facility is currently provided by the European Community's Seventh Framework Programme Biostruct-X. For further information, please contact spc@emblhamburg.de.





Keywords: EMBL@PETRAIII synchrotron facilities, High-throughput thermofluor and crystallization, Sample preparation and biophysical characterization