



	Experiment title: Investigation of the initial stage of protein crystallization by SAXS	Experiment number: MX-2089
Beamline: BM29	Date of experiment: from: 29.07.2018 to: 30.07.2018	Date of report: 29.10.2018
Shifts: 3	Local contact(s): Mark Tully	<i>Received at ESRF:</i>
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Report:

The thermolysin solutions were studied with and without a precipitant addition at several protein and precipitant concentrations and temperatures of 20, 15 and 10 °C by small-angle X-ray scattering. In addition to the expected protein monomers, the dimeric and the hexameric species were identified in solutions upon addition of the precipitant (Figure 1). In contrast, the system of attractive monomers cannot adequately describe experimental data. The volume fraction of the hexamers increases while the precipitant concentration increasing and the temperature decreasing. The selection of the hexamer as a base crystal growth unit for the hexagonal thermolysin was supported by its crystal structure analysis.

The article “Pre-crystallization phase Formation of thermolysin hexamers in solution close to crystallization conditions” is in press (Journal of biomolecular structure & dynamics).

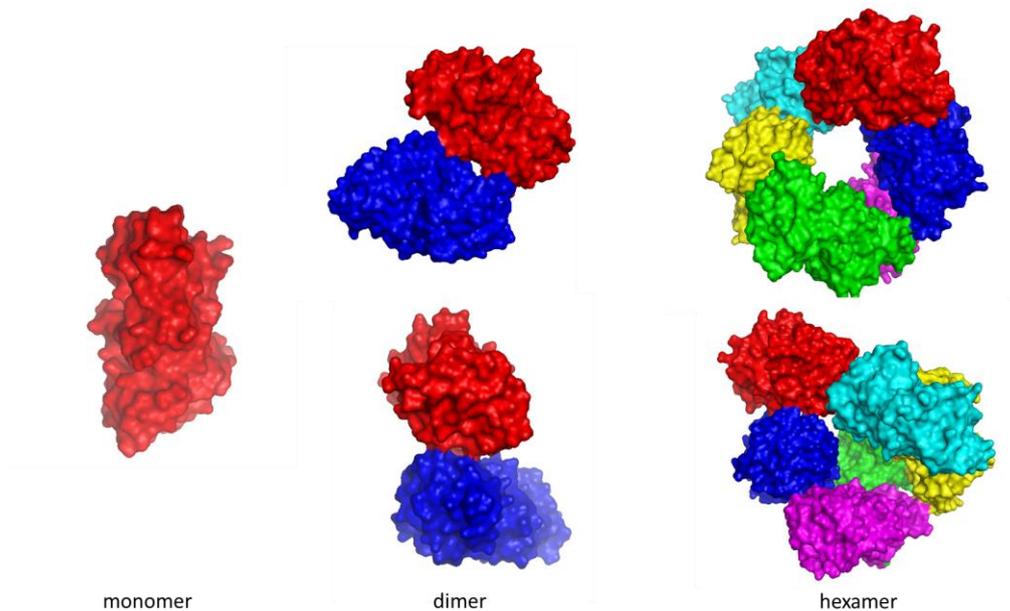


Figure. 1. Oligomers of thermolysin used for the analysis of SAXS data. For clarity, the thermolysin molecules are displayed in different colors. The image was created using PyMol software.

The composition of glucose isomerase solutions under conditions favorable for the formation of orthorhombic crystals has been investigated at different protein concentrations by small-angle X-ray scattering. The glucose isomerase solutions were studied with and without a precipitant addition at several protein and precipitant concentrations and temperatures of 20 °C. In addition to glucose isomerase monomers, dimeric, tetrameric and octameric species are found in the crystallization solutions; the octamer content increases with an increase in the protein concentration (Figure. 1.). SAXS study showed that tetramers and octamers are formed in the initial stage of glucose isomerase crystal growth and that the volume fraction of octamers increases with an increase in the protein concentration.

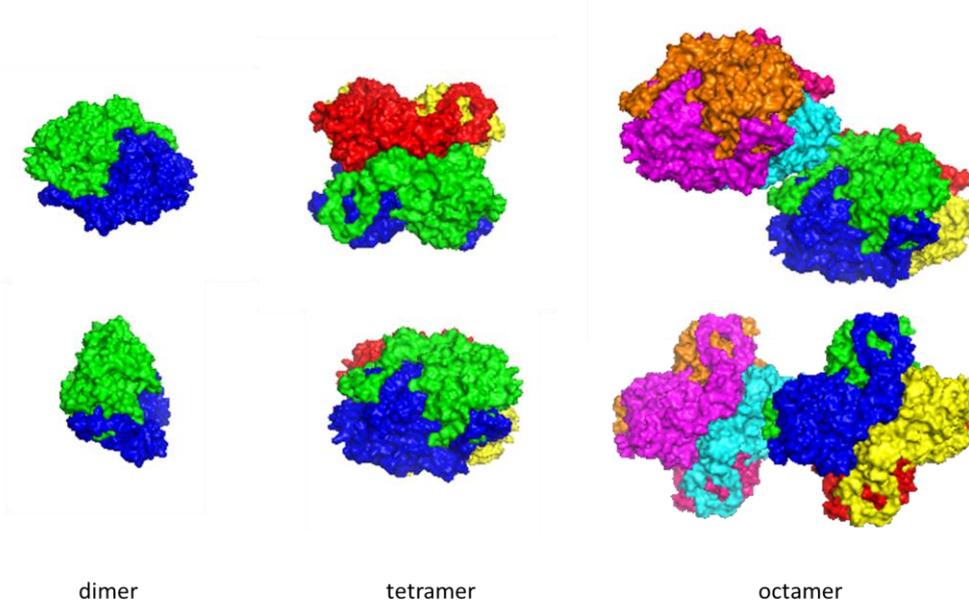


Figure. 2. Oligomers of glucose isomerase used for the analysis of SAXS data. For clarity, the glucose isomerase molecules are displayed in different colors. The image was created using PyMol software.