

116c Determination of Protein Crystallization Kinetics Using a Regulated Evaporation Method

Sameer Talreja, Charles F. Zukoski, and Paul J. A. Kenis

Proteins are difficult to crystallize and the current methods rely on high throughput screening methods, in which hundreds to thousands of solution conditions are screened to locate and optimize the solution conditions where X-ray diffraction quality crystals are formed. We have developed a method that ensures the formation of a solid phase in each experiment and, as a result, each trial yields information on crystal solubility, and crystal nucleation/growth rates. This method relies on evaporating the solvent from a drop at a predetermined rate set by the dimensions of the crystallization platform. Depending on the initial protein and precipitant concentrations and the rate of evaporation, this method yields different solid phases, including amorphous precipitates, gels, showers of small crystals, a few large crystals, or combinations of those. Once solution conditions resulting in good crystals are found, monitoring the crystal size during the experiment provides a way to extract physico-chemical parameters of importance to crystal growth. We use hen egg-white lysozyme as the model protein in this study. The experiments are highly reproducible in number of crystals formed and growth rates. We apply a population balance model to the experimental data to extract the kinetic parameters for crystal nucleation and growth and compare these values with data from literature for lysozyme. The experimental data of number of crystals and growth rate are in excellent agreement with the predictions of the model.