4x Microfluidic Devices for Protein and Pharmaceutical Crystallization Applications

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My research is geared towards developing the fundamental understanding of the crystallization processes in protein and pharmaceutical systems. This knowledge will greatly help not only the practice of crystallization of proteins for structure determination, but also rational drug design methodology and controlling of pharmaceutical production processes. Three specific problems that will be covered in this poster are:

1. Identifying Key Solution Parameters in Protein Crystal Nucleation

Crystallization of biological molecules is the first step (and usually a rate limiting step) in structure determination through X-ray diffraction. Formation of stable crystals of proteins and protein/nucleic acid complexes from solution is dominated by the kinetics of nucleation of these clusters. Given the sparse amount of kinetic data available, the nucleation rates of a model protein system, tetragonal hen egg-white lysozyme (HEWL), were measured at various solution conditions by the method of initial rates. These data were modeled using an empirical kinetic expression based on the classical nucleation theory. Also, it was observed that, for HEWL / NaCl system, equal solubility conditions produce equal nucleation kinetics at a given initial protein concentration, even when the solution conditions such as pH, ionic strength and temperature are different. This observation, if shown to be valid for all protein systems, could have far reaching consequences, as it will significantly narrow the spectrum of parameters to be explored to identify optimal nucleation and crystallization conditions.

2. Estimation of Crystallization Kinetic Parameters of Pharmaceutical Compounds using Regulated Evaporation of a Microdrop

Crystallization of pharmaceutical compounds from solution is usually limited by inadequate knowledge of nucleation and crystal growth kinetics. To address this issue, an evaporation-based method is developed. This method facilitates rapid generation of experimental data useful in understanding crystallization processes. Solution droplets at various initial conditions are introduced into a microdevice, in which crystallization is driven by different rates of evaporation of the solvent. The droplets are observed periodically using an optical microscope and the nucleation time for any crystals formed is recorded. Metastable zone limits for several systems for a wide range of initial conditions were obtained. This metastable limit was observed to be independent of the rate of solvent evaporation, i.e. the rate of change of supersaturation. Several model systems, ranging from amino acids to bio-macromolecules, exhibit the same behavior. Furthermore, it was observed that this limit is strongly correlated to the solubility for different chemical compounds.

3. High Throughput Polymorph Screening using Microfluidic Devices

A significant interest exists in the pharmaceutical industry for rapid polymorph screening techniques that reduce the time, effort and material consumed. Towards that end, we are developing a microfluidic solution for high throughput polymorph screening. Here, a ‘first generation' microfluidic mixer design is discussed. This design effects anti-solvent crystallization utilizing the fact that mixing at the microscale is by diffusion only (no turbulence). Also, the rate of mixing and the composition of the liquid phase can be varied by changing the flow rates and concentrations, thus influencing the polymorph selectivity. The results of our experiments on some model systems are presented and the applicability of the concept towards the development of a high throughput microfluidic device for polymorph screening are discussed.