



Supporting Information

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**A Droplet-based, Composite PDMS / Glass Capillary Microfluidic System for Evaluating Protein
Crystallization Conditions by Microbatch and Vapor Diffusion Methods with On-Chip X-Ray
Diffraction**

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Fabrication of Devices: We followed the procedure described in Langmuir 2003, 19, 9127 with the following modifications. The microfluidic device in our system consisted of two parts: the first part is a polydimethylsiloxane (PDMS) microfluidic with channels of $200 \times 200 \mu\text{m}^2$ cross-sectional dimensions; the second part is an x-ray capillary with ID $180 \mu\text{m}$ and OD $200 \mu\text{m}$ (Hampton Research). The capillary was cleaned by chromic acid then treated with (tridecafluoro-1,1,2,2-tetrahydrooctyl)-1-trichlorosilane (United Chemical Technologies, Inc.) for at least 30 min, rendering the inner surface of the capillary hydrophobic. The capillary was connected to the outlet of the PDMS device and the junction was sealed by partially cured PDMS. The partially cured PDMS was allowed to fully cure immediately afterwards in a 60°C oven.

Microbatch Crystallization in Capillaries: Stock solutions of 50 mg/ml thaumatin (Sigma) in 0.05 M pH 6.5 buffer of N-(2-acetamido)iminodiacetic acid and precipitant (1.0 M sodium potassium tetrates in 0.03 M pH 7.5 buffer of N-[2-hydroxyethyl]piperazine-N'-[2-ethenesulfonic acid]) were used.

Perfluoroperhydrophenanthrene (Acros Organics) with 10 % (v/v) 1H,1H,2H,2H-perfluoro-1-octanol (Acros Organics) was used as oil. Droplets of mixture of thaumatin and precipitant at volume ratio 1:1 were formed in the PDMS channel and were carried into the capillary by the flow of oil. After the capillary was filled with droplets, the flows were stopped and the capillary was cut off and sealed with wax. The capillary was incubated at 18°C and monitored periodically under a microscope.

Vapor Diffusion Crystallization in Capillaries: Stock solutions of 75 mg/ml lysozyme (Sigma) in 0.05 M NaAc pH 4.5 buffer, precipitant (25 % w/v PEG 5000, 1.0 M NaCl in 0.05 M NaAc pH 4.5 buffer) and high-concentration salt solution (15 % w/v PEG 5000 and 4.0 M NaCl) were used.

Polytrifluoropropylmethylsiloxane homopolymer (FMS-121, from GELEST, Inc.) saturated with 1H,1H,2H,2H-perfluoro-1-octanol (Acros Organics) at 4°C was used as oil. Droplets with alternating

compositions were formed in the PDMS channel, and were carried into the capillary by the flow of oil. After the capillary was filled with protein/salt droplet pairs, the flows were stopped and the capillary was cut off and sealed with wax. Size change of droplets and crystallization of protein were monitored periodically under a microscope.

On-Chip X-Ray Diffraction: X-ray diffraction was performed at BioCARS station 14BM-C and 14ID-B at the Advanced Photon Source at Argonne National Laboratory. Beam wavelength was 0.90 Å for thaumatin and 0.83 Å for lysozyme, respectively. Capillaries were cut to the appropriate length without disturbing crystal-containing droplets, resealed using capillary wax, and mounted on clay-tipped cryoloop holders at a distance of 12 ± 5 mm from base to crystal. The holder was placed on the x-ray goniometer. Crystals were centered on the beam. Snapshots were taken using 10 seconds (thaumatin) and 5 seconds (lysozyme) exposures. Distance from sample to detector was 150 mm. Snapshots were analyzed using the HKL/Denzo analysis package.