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Aggregated Pre-Crystallization State of Ferritin seen by Solution Scattering at Low Temperature. S. Boutet, I.K. Robinson, Dept. of Physics, Univ. of Illinois at Urbana-Champaign, 1110 West Green St., Urbana, IL, 61801.

In the course of a systematic exploration of the crystallization kinetics and conditions of the protein ferritin using x-rays, we discovered an unexpected new state of aggregation of the protein at low temperature. This new state was found to form reversibly below the freezing point of the solution. Using Small Angle X-ray Scattering (SAXS) which allows one to probe the internal structure of solutions, we studied the properties of solutions of ferritin upon cooling.

We found that ferritin in solutions not containing the cadmium salt usually needed for crystallization, forms aggregates upon freezing. This is evidenced by a strong broad peak observed in the SAXS pattern at a value of momentum transfer corresponding roughly but not exactly to the first crystal Bragg peak of ferritin. Such aggregates are thus neither crystalline nor fully amorphous. Furthermore, this peak is seen to move with temperature, which corresponds to a change in the average spacing of the protein molecules. From this, we obtained a surprisingly large thermal expansion of roughly 30% over a range of 30˚C. The freezing process was also found not to destroy preexisting crystals in solutions containing cadmium salt. Upon freezing, these crystals were seen to remain while the broad peak from the new aggregated state was seen to appear. This state and the crystalline state can coexist. This leads us to propose that this state may be a precursor state to crystallization. We will present SAXS data on solutions of the 2 common forms of ferritin: apoferritn which contains no iron and holoferritin which has a large iron core.