

## Effects of Salts on Protein Aggregation and Crystallization

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### Summary

A structure analysis of proteins provides fundamental and useful information for the research on biochemistry and life science and for the development of new medicines and functional foods. The protein crystallography needs good crystals for suitable to x-ray or neutron diffraction. However, crystallization of proteins is a bottleneck because it is a try and error procedure and requires considerable experimental efforts.

We have found that a forward light scattering at small angles ( $< 8^\circ$ ) is highly sensitive to aggregations and crystal nucleus formation of proteins in crystallizing protein solutions <sup>[1]</sup>. Previous work <sup>[2]</sup> has shown that crystallizing solutions of Hen-Egg-White Lysozyme (HEWL) with NaCl have a forward static light scattering (F-SLS) pattern of non-integer power law, which indicates a formation of fractal aggregates.

Here, we evaluated an effect of precipitant salts of NaCl, KCl, MgCl<sub>2</sub>, and CaCl<sub>2</sub> with chlorine ion, and Na<sub>2</sub>SO<sub>4</sub> salt with sulfate ion on the HEWL crystallization, using *in-situ* detection technique for F-SLS measurements. We succeeded observation of the changeable aggregation by the added salts in pre-crystalline HEWL solutions. A formation of the protein fractal aggregates with relatively high density requires some time, for example, over twenty minutes in the crystallizing solutions.

[1] T. Wakamatsu, "Forward light scattering for highly sensitive detection of aggregation in crystallizing protein solutions," *Applied Physics Letters*, **98**, 263701, 2011.

[2] T. Wakamatsu, "Forward-Light-Scattering Characterization of Pre-crystalline Aggregates in Crystallizing Lysozyme Solutions," *American Journal of Analytical Chemistry*, **5**, 581, 2014.