

agents against Gram-positive pathogens. Here we report new binding and structural studies of FapR from *Staphylococcus aureus* in complex with both its DNA operator and effector molecules, providing important hints to understand the mode of action of this conserved bacterial repressor.

Keywords: protein-DNA interactions, regulation of fatty acid biosynthesis, X-ray crystallography

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Seeing the first stages of protein crystal nucleation through to a full powder pattern

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The nucleation and growth of protein crystals is the crucial process in any protein structure analysis. Powder diffraction provides a possible means for study of this process because crystallinity can readily be established by diffraction, and a time series of diffraction patterns can be used to explore the crystal growth. Analysis of this data shows not only the time dependence of the amount of crystallized material but also the dimensions of both the crystallites and the crystal unit cell. This talk will show an example of the crystallization of hen egg white lysozyme examined in real time by powder diffraction.

Keywords: powder crystallography, protein crystallization, kinetics of growth

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Serial crystallography: Use of a micro-jet for diffraction of protein nano-crystals or molecules

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We propose a method of acquiring protein powder diffraction data from the smallest available protein crystals that can be acquired through batch precipitation. A continuous micro-jet is used to deliver a solution of hydrated protein nano-crystals to an x-ray beam. This method requires neither the crushing of larger polycrystalline samples nor any techniques to avoid radiation damage such as cryo-cooling. Radiation damage is completely avoided because of the very small dose each crystallite receives as it is only briefly exposed to x-rays. We have commissioned an apparatus to record protein powder diffraction in this manner and in this talk present the first such patterns from photosystem-1 crystals with sizes less than 500 nm. These preliminary patterns show the lowest order reflections, which agree quantitatively with theoretical calculations of the powder profile. The results also serve to test our flow-focusing arojet injector system, with future application to femtosecond diffraction in Free Electron X-ray Laser schemes, and for Serial Crystallography using a single-file beam of aligned hydrated molecules.

Keywords: serial crystallography, nanocrystals, radiation damage

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Femtosecond laser etching of protein crystal to process and to isolate the single crystal

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Preparation technique of single protein crystal with suitable shape is needed for precise X-ray diffraction (XRD) measurement giving three-dimensional structure of protein. We have applied an infrared femtosecond laser to isolate and process protein crystal with irregular shape. When the intense femtosecond laser is focused on the protein crystal, electronically excited state of protein molecule is generated densely and the energy is distributed to vibrationally excited states of the molecules in time scale shorter than ns. On the other hand, the time scale for the morphological change of the crystal is longer than ns. Because of the time gap between the photoexcitation energy distribution and the morphological change, a stress is confined at the laser focal point in the time scale shorter than ns. As the result, the etching of the protein crystal by the femtosecond laser is initiated mechanically by the stress increase, though the etching by the other lasers is generally attributed to explosive sublimation explained by conventional heat generation process. The excellence of the femtosecond laser etching is that the cutting is enhanced by a cleaving behavior resulting to stress propagation from the laser focal point. Conclusively the protein crystal has hardly thermal damage by the femtosecond laser irradiation, which was checked by XRD measurement of the small cut component. Furthermore, debris of protein crystal generated by the femtosecond laser etching can be utilized as seed crystal. We confirmed that crystallinity of daughter crystal grew from the seed crystal was fine or comparable in comparison with the mother crystal. These techniques are applicable to select a single crystal from multiple growth forms, even when the size is micrometer-order.

Keywords: protein crystals, pulsed laser ablation, femtosecond laser

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Features of the secondary structure of protein molecules from powder diffraction data

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The determination of crystal structures greatly depends upon the availability of good quality single crystals, the growth of which is currently one of the major bottlenecks in macromolecular