MS36 O4

Quantifying X-ray Radiation Damage in Protein Crystals Robert E. Thorne,[a] Jan Kmetko[b], Matthew Warkentin[a], and Ulrich Englich[c]; *[a]Physics Department, Cornell University, Ithaca, NY 14853, [b]Physics Department, Kenyon College, Gambier, OH 43022, [c]MacCHESS, Cornell University, Ithaca, NY 14853*

Keywords: protein crystallography; radiation damage; cryocrystallography

We have examined how radiation damage to protein crystals at cryogenic temperatures depends upon the crystal's X-ray absorption cross-section [1]. Lysozyme crystals containing varying heavy atom concentrations were irradiated, and diffraction patterns recorded as a function of the total number of incident photons. We define an experimental protocol that monitors degradation of the relative, isotropic B-factor and a coefficient of sensitivity to absorbed dose, and show that together they yield a sensitive and robust measure of damage. Radiation damage per incident photon increases linearly with the crystal's absorption coefficient, but damage per absorbed photon is the same for all heavy atom concentrations. Using this protocol and damage metric, we have examined crystals of five proteins with a wide range of molecular weights, solvent contents and room-temperature radiation sensitivities. At cryogenic temperatures, all five show similar damage per absorbed photon. X-ray radiation sensitivity of protein crystals — properly defined — may thus be roughly independent of the crystal composition at cryogenic temperatures, consistent with results in electron diffraction. This will simplify automated optimization of data collection protocols to minimize X-ray damage. We have also examined the effects of a wide variety of free radical scavengers on radiation sensitivity of simple model proteins at room and cryogenic temperatures. Only one — nitrate ion — shows protective effects at room temperature, and none show protective effects at cryogenic temperatures.

[1] J. Kmetko *et al*., Acta Cryst. D 62, 1030–1038 (2006).

MS36 O5

Phasing in the Home Laboratory Richard Tyrrell, Joseph D. Ferrara, Cheng Yang, Robert Bolotovsky, James W. Pflugrath Rigaku Americas Corp., The Woodlands, TX, 77381

Keywords: SAD, phasing, chromium

Many examples of S-SAD and Se-SAD phasing have been reported with diffraction data collected using copper radiation (1.54 Å) or radiation at the selenium K absorption edge (0.98 Å). With recent advances in X-ray technology, chromium radiation (2.29 Å) is now available for in-house data collection and appears to be ideally suited for measuring anomalous signals from weak anomalous scatterers such as sulfur, selenium, calcium and other atoms commonly found in protein crystals. The results of a number of successful SAD experiments using Cr radiation have been published by several groups including our own.

With the addition of Cr radiation to the crystallographer's toolkit, in-house X-ray sources can provide at least two routinely useful wavelength options for macromolecular crystallography. This report also discusses the results of phasing by combining diffraction data collected using both Cu and Cr radiation sources.

Finally, we report the results of data collection with a new imaging plate detector (R-AXIS HR) designed specifically for use with Cr radiation. This new detector allows the collection of data suitable for both phasing and refinement with Cr radiation from a single crystal in a single, simple diffraction experiment.