Experimental Tests of the Theoretical Dose Limit for Cryocooled Protein Crystals

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Various aspects of radiation damage to protein crystals at 3rd generation synchrotron sources have been investigated. Experiments have been carried out on beamline ID14-4 at the ESRF to investigate the effect of the dose, dose-rate, and the macromolecular composition on the degradation in diffraction by, and derived structure of, apoferritin and holoferritin crystals.

To compare with the theoretical dose limit of 2×10^7 Gy [1] for half the diffraction pattern to disappear, the dose absorbed by a crystal must be calculated. This is a function of both the beam parameters and the crystal composition and can be determined using RADDOSE [2]. The elemental composition was measured using microPIXE [3].

The decay in diffraction of four holoferritin and three apoferritin crystals was analysed. Crystals were exposed to an unattenuated beam between datasets; changes in diffraction quality and crystal structure could then be analysed as a function of the cumulative absorbed dose.

A new dose limit for cryocooled protein crystals is proposed and linked to experimentally observed reductions in diffractive and structural quality.

[1] Henderson R., *Proc. R. Soc. Lond. B*., 1990, **241**, 6. [2] Murray J. W., Garman E., Ravelli R., *J. Appl. Cryst.*, 2004, **37**, 513. [3] Garman E., *Structure,* 1999, **7**, R291.

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