

MS47-02 Systematic characterization of radiation sensitivity and its applications for MX data collection. Gleb Bourenkov,^a Ricardo F. Leal,^b Silvia Russi,^b Alexander N. Popov,^b ^aEMBL-Hamburg Outstation, Notkestrasse 85, 22607 Hamburg, Germany, ^bESRF, Rue Jules Horowitz, BP220, Grenoble, France
E-mail: gleb.bourenkov@embl-hamburg.de

Given an approximate knowledge of radiation sensitivity of macromolecular crystal expressed in a suitable quantitative metrics related with the diffracting power of a crystal, optimal data collection strategy can be devised within a systematic approach implemented in software BEST [1]. The problem of calibrating the radiation sensitivities at cryo- and room-temperatures using a “sacrificial sample” approach is now implemented at ESRF MX beamlines as a fully automated procedure [2]. The protocols developed are robust and reliable, so far that they are also being employed for the beamline diagnostics and beam quality assurance. We evaluate variation of the overall scale and isotropic temperature factors with the dose using two correlated parameters; these are directly used in the strategy optimization. With appropriate corrections for instrumental factors, like intensity variation across the incident beam profile, the model fits the experimental data very accurately; equivalent representation of the model via a single sensitivity parameter $D_{1/2}$ [3] is used as an absolute metrics. Experimental data accumulated on a substantial number of structures at cryo temperatures suggest essentially low, <30%, variation among the different structures. Recently we carried out the survey of radiation sensitivities on large selection of the model structures at room temperature using open-flow humidity control device HC1b [4]. As compared to cryo-temperatures, where variation in overall B-factor dominates, at room temperature significant component associated with the overall scale is clearly present. The sensitivities vary between different structures by orders of magnitude. The data confidently show (the only) systematic dependency of the sensitivity on the crystal solvent content. No dose-rate dependency is observed over a broad range of dose rates, 0.5 to 500 kGy/sec.

- [1] Bourenkov, G.P. & Popov, A.N. (2010). *Acta Cryst.* **D66**, 409-419.
- [2] Leal, R.F.M., Bourenkov, G.P., Svensson, O., Guijarro, M. & Popov, A. (2011). *J. Synchrotron Rad* **18**, 381-386.
- [3] Owen R.L., Rudino-Pinera, E., Garman E.F. (2006), *PNAS* **103**, 4912-4917
- [4] Sanchez-Weatherby, J., Bowler, M.W., Huet, J., Gobbo, A., Felisaz, F., Lavault, B., Moya, R., Kadlec, J., Ravelli, R.B. & Cipriani, F. (2009). *Acta Cryst.* **D65**, 1237-1244.

Keywords: radiation damage; radiation sensitivity metrics, data collection strategy

MS47-03 Serial femtosecond crystallography: structure determination and radiation damage analysis. Lukas Lomb,^{a,b} ^aMax-Planck-Institute for Medical Research, Heidelberg, Germany, ^bMax-Planck Advanced Study Group, CFEL Hamburg, Germany,
E-mail: Lukas.Lomb@mpimf-heidelberg.mpg.de

Elucidation of the structure of proteins without being affected by radiation damage is a dream of structural biologists. Data collection using free-electron lasers promises to allow a significantly increase in the dose tolerated by protein crystals, relieving requirements on crystal size.

Serial femtosecond crystallography is a recently established method [1] to collect diffraction data using a stream of (sub-)micron sized crystals exposed to high-intensity femtosecond pulses of an FEL.

Irradiating protein crystals with up to 10^{13} 2 keV photons per femtosecond pulse at the AMO beamline of the LCLS facility [2], radiation damage has been studied for doses exceeding 100 times the conventional dose limit of 30 MGy [3] using the model systems PSI [4] and lysozyme [5].

Variation of the FEL pulse lengths revealed increasing radiation damage for pulse lengths exceeding 100 fs. A pulse length internal classification of the diffraction patterns showed a damage dependence on the incident irradiance. Even for the shortest pulses (70 fs) used, signs of radiation damage were observed. Although rescaling the data sets allows accounting for an increased homogeneous decrease of the crystalline order leading to self-termination of the Bragg peaks [4], scaling the data by a linear k-factor an overall B-factor cannot account for all observed changes in structure factor intensities [5].

Data sets of lysozyme crystals, collected at the CXI beamline at LCLS using 9.4 keV photons with pulse lengths of 5 fs and 40 fs at doses comparable to the conventional dose limit, show very good statistics and compare well to conventional data sets to a resolution of 1.9 Å.

The described experiments were carried out by “the Imaging Collaboration” with contributions from the Max-Planck-Institute for Medical Research, the Max-Planck Advanced Study Group (ASG), CFEL DESY, Arizona State University, SLAC, University of Hamburg, Uppsala University, LLNL and LBNL.

- [1] H. Chapman et al., *Nature (London)* **470**, 73 (2011)
- [2] P. Emma et al., *Nature Photonics* **4**, 641 (2010)
- [3] R. Owen, E. Rudino-Pinera, and E. Garman, *Proc. Natl. Acad. Sci USA* **103**, 4912 (2006)
- [4] A. Barty et al., *Nature Photonics* **6**, 35-40 (2011)
- [5] L. Lomb et al., *Physical Review B* **84**, 214111 (2011)

Keywords: x-ray free-electron laser; serial femtosecond crystallography; radiation damage