Consideration of improved data accuracy in solvent removal and crystal processing using the deep UV laser

Ayaka Harada [1], Naohiro Matsugaki [1],Masahide Hikita[1], Toshiya Senda [1]

[1]Structure Biology Research Center, Institute of Materials Structure Science, KEK

Email Contact: a.harada0509@gmail.com

Native SAD phasing uses anomalous scattering signals from light atoms such as sulfur and phosphorus in protein crystals. The anomalous signals from these atoms are, however, much weaker than those from heavy atoms that are frequently utilized in protein crystallography. Therefore, high-quality data collection is essential for native SAD phasing. Lower energy X-ray is favorable to enhance the anomalous signals but on the other hand, X-ray absorption by sample, solvent, and air hinder the high-quality diffraction data collection. Here, we present a method minimizing the X-ray absorption; the solvent portion of a mounted frozen crystal is removed or the mounted crystal is spherically shaped by the deep UV laser processing technique, which was developed in SPring-8 (Kitano, 2004), (Murakami, 2004), (Basu, 2019). A previous study has revealed that data statistics of diffraction data that were collected using X-ray of 3.7 keV at BL-1A of Photon Factory (Tsukuba, Japan) were significantly improved when the crystals were processed by the deep UV leaser.

In this study, crystals of BphA4 and LigM proteins were used. Their theoretical bijvoet ratios are 1.69 % and 1.98 % at the X-ray of 3.7 keV, respectively. Using deep UV leaser, we processed crystals of BphA4 and LigM, of which crystal structures were already determined. The diffraction data were collected using the X-ray of 3.7 keV at BL-1A. All diffraction data were collected with a helium chamber to reduce X-ray absorption by air. Each data was processed and scaled by XDS and XSCALE, respectively (Kabsch, 2010). The obtained data were used for native SAD phasing with autoSHARP (Vonrhein, 2007). In this presentation, we will show the results of native SAD phasing with deep UV processed/unprocessed crystals.