Crystallography

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Assessment of Perfection of Hen Egg-white Lysozyme Crystals by X-ray Digital Topography with CCD Camera

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Introduction

X-ray diffraction techniques such as X-ray topography and rocking curve measurement are powerful methods for the characterization of crystal defects especially dislocations and evaluation of crystal perfection. The application of X-ray topography and rocking curve measurement to protein crystals have been carried out by a lot of groups [1]. For the comprehensive understanding of crystal perfection of protein, the same crystal needs to be evaluated by both X-ray topography and rocking curve measurement.

Recent development of high-resolution X-ray CCD camera enables to higher spatial observation. By using the CCD camera, X-ray topographic images are successively recorded with the rotation of the specimen as the rocking curve measurement. This method with CCD camera can lead to the simultaneous observation of X-ray topographic images and local rocking curves. In addition, the shorter exposure with CCD camera is effective especially for protein crystals, since they easily suffer some damages by X-ray irradiation. In this paper, we report the evaluation of perfection of protein crystals by X-ray digital topography.

Experiments

Tetragonal hen egg white lysozyme (HEWL) crystals were grown by the salt concentration-gradient method as reported elsewhere [1]. These crystals were used for X-ray topographic experiments.

X-ray digital topography was carried out in BL15B1 and BL15C1. The monochromatic-beam of 1.2 Å was selected by adjusting the double-crystal monochromator. The topographs were taken by the synchrotron monochromatic beam which was almost perpendicular to the (0 0 1) plane of tetragonal HEW lysozyme crystals. The topographs were recorded on high-resolution X-ray CCD camera (Photonic Science XFDJ). The camera length was 25 cm. The digital topographic images were recorded at the interval of 0.001 degrees. The size of one pixel in CCD is 6.4×6.4μm^2.

In order to handle the large amount data of the sequential digital topography images, we developed appropriate software program [2]. The peak analysis of local rocking curve at one pixel was performed by our program. The angular position of the peak top and the full width of half maximum (FWHM) of the peak were estimated.

Results and Discussion

The left picture in Fig. 1 shows X-ray digital topographic image taken with CCD camera. The right upper figure shows a local rocking curve taken at the position (one pixel) marked A which is included in the perfect region in the topograph. The right bottom figure shows another local rocking curve taken at the position marked B which is included in the dislocation image in the topograph. The rocking curve at A exhibit single peak with narrow FWHM of 0.0016 degree. On the other hand, the rocking curve at B exhibit double peak. Therefore, the peak profile strongly depend on the crystal perfection or defect character. Thus, these analyses will lead to more detailed understanding of perfection in protein crystal.

References

[1] T. Sawaura et al. J. Crystal Growth **318**, 1071 (2011)
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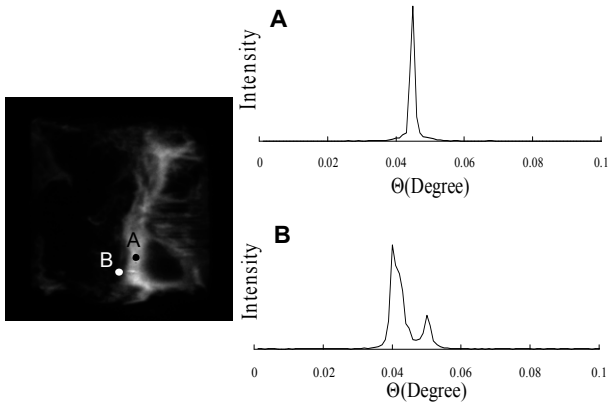

Fig.1 X-ray digital topographic image and the corresponding local rocking curves which were taken at the positions (one pixel) marked A and B in the topography, respectively.