Challenges to high-throughput protein micro-crystallography at SPring-8

Masaki Yamamoto*[1], Keitaro Yamashita[1], Kunio Hirata[1], Kazuya Hasegawa[1,2], and Takashi Kumasaka[1,2]

[1]*RIKEN SPring-8 Center, Japan*

[2]*JASRI/SPring-8, Japan*

[*]*yamamoto@riken.jp*

Macromolecular crystallography coupled with a highly brilliant X-ray beam from advanced SR and XFEL sources along with novel data collection and processing, continues to shed light on many challenging and scientifically important problems. The crystallographic analyses of many challenging and scientifically important proteins are still difficult due to their small and weakly diffracting crystals. The modern micro-focus beamlines providing μm-order high brilliant beam are suitable for data collection from small and weakly diffracting crystals including membrane proteins [1]. BL32XU at SPring-8 is a beamline dedicated to protein micro-crystallography [2]. We have developed a high-throughput data collection pipeline named ZOO for obtaining datasets from any types of crystals. For microcrystals, the small-wedge mode of ZOO realized fast raster-scan to locate crystal positions and the subsequent small-wedge (5-10°) shutterless data collection with a high-speed detector, and the data were processed by the pipeline program KAMO [3]. The whole process from sample exchange to data processing was automated in ZOO system, which has contributed to many structure analyses from many microcrystals including membrane proteins. On the other hand, it is sometimes difficult to find smaller micro-crystals by raster scan and collect data due to radiation damage. We examined a new one-step approach to obtain high-resolution data from smaller micro-crystals inspired by the Gati's protocol [4]. The approach is 'fixed target serial synchrotron rotation crystallography (SS-ROX)' [5]; multiple diffraction images are collected with the shutterless helical raster scan using plenty of micro-crystals loaded on a loop. In order to find the optimum data collection condition without the fatal radiation damage, we collected SAD data by this method at BL41XU. Data statistics and electron density map were evaluated using the data collected with various oscillation ranges and radiation doses.

We present the current status and the future of protein micro-crystallography at SPring-8.

References

[1] Yamamoto, M. *et al.* (2017) *IUCrJ* **5**, 529-539

[2] Hirata, K. *et al.* (2013) *J. Physics: Conf. Series* **425**, 012002

[3] Yamashita, K. *et al.* (2018) *in preparation.*

[4] Gati, C. *et al.* (2014). *IUCrJ*. **1**, 87–94.

[5] Hasegawa. K. *et al.* (2017) *J. Synchotron Rad.* **24**, 29–41.