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Cryogenic coherent X-ray diffraction imaging of biological particles at SACLA

Y. Takayama[1], M. Nakasako[1,2], T. Oroguchi[1,2], Y. Sekiguchi[1,2], M. Yamamoto[1], K. Yonekura[1], Y. Takahashi[1,3], A. Suzuki[1,3], S. Matsunaga[4], Y. Tsujimoto-Inui[4]

[1]RIKEN SPring-8 Center, Sayo, Hyogo, Japan, [2]Keio University, Department of Physics, Yokohama, Kanagawa, Japan, [3]Osaka University, Department of Precision Science and Technology, Suita, Osaka, Japan, [4]Tokyo University of Science, Department of Applied Biological Science, Noda, Chiba, Japan

Coherent X-ray diffraction imaging (CXDI) is a promising technique to visualize internal structures of whole biological cells without sectioning. Utilizing X-ray free-electron laser (XFEL) for CXDI has potential to collect huge number of projected electron densities of such samples at higher resolutions than that limited by radiation damage. For biological application of XFEL-CXDI, sample particles must be kept in a hydrated state to maintain their functional structures, but be placed in vacuum to obtain weak diffraction signals. In addition, we need to deliver fresh sample particles one after another for XFEL exposure because every particle explodes just after X-ray irradiation. To handle these problems, we have been developing a system to fulfill cryogenic XFEL-CXDI of frozen-hydrated specimens. For cryogenic XFEL-CXDI, we prepare frozen-hydrated specimens by plunge-freezing sample particles dispersed onto thin film with humidity-controlling [1]. The diffraction experiments are conducted by using the cryogenic X-ray diffractometer KOTOBUKI-1 [2]. In a vacuum chamber of KOTOBUKI-1, a cryogenic pot equipped on a goniometer is filled with liquid nitrogen, which cools the specimen via thermal contact. Thus, KOTOBUKI-1 allows data collection at a specimen temperature of ~66 K with a positional fluctuation of less than 0.4 μm. A small angle-resolution of better than 500 nm is attainable by using a pair of L-shaped Si-slits placed before the specimen, which eliminate almost all parasite scattering from upstream. Diffraction patterns recorded on two MPCCD detectors in tandem arrangement are automatically processed and phase-retrieved by program suite SITENNO [3]. In our recent experiments performed in Japanese XFEL facility SACLA, we were able to collect a large number of diffraction patterns from biological samples at a resolution of 50 - 30 nm at an XFEL hit-rate of 20 - 100%. We report details of the cryogenic XFEL-CXDI and introduce imaging of chloroplasts as an example.

[1] *Y. Takayama & M. Nakasako, Rev. Sci. Instrum., 2012, 83, 054301 (6 pages).,* ***[2]*** *M. Nakasako, Y. Takayama, T. Oroguchi et al., Rev. Sci. Instrum., 2013, 84, 093705 (11 pages).,* ***[3]*** *Y. Sekiguchi, T. Oroguchi, Y. Takayama et al., J. Synchrotron Rad., in press*

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