

3rd Workshop on the Simultaneous Combination of Spectroscopies with X-ray Absorption, Scattering and Diffraction Techniques



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Present status of micro-spectroscopic instruments in SPring-8 macromolecular crystallography beamlines

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Spectroscopic measurement combined with X-ray diffraction study is an important technique to elucidate the structure-function relationships of proteins. At SPring-8, two types of on-line micro-spectrophotometer (MS) for absorption spectroscopy have been dedicated to macromolecular crystallography (MX). The one is designed to measure Vis absorption spectra simply and at high-speed with CCD array detector installed at BL26B2. Another type has features of high-sensitivity and wide dynamic range by photomultiplier and monochromatic light source which covers UV-Vis region in BL38B1. Both have been used for validation of radiation damage or protein cofactor state (e.g. photoreceptor, oxidation-reduction, etc.).

As our study using MS, radiation damage against protein structure was evaluated by absorption spectroscopy at BL38B1. As the results, decrease of the absorption maximum at UV region was observed, and it indicates the state change of aromatic residue, in spite of only modest change of electron density map.

We are now planning to build a new on-line MS at the MX beamlines in SPring-8. This is expected to target the UV-Vis absorption and Raman spectroscopy for precise evaluation of radiation damage, detailed validation of cofactor state, and detection of entrapped reaction intermediates on protein crystals. It hopefully performs simultaneous measurement with diffraction data collection.

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