

Stability of solution structure and photo-induced structural changes of photoactive yellow protein with use of mutants

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Photoactive yellow protein (PYP) is present in *Ectothiorhodospira halophila* as a soluble small chromoprotein. It has a broad absorption band, the absorption maximum of which is at 446 nm. It is considered a photoreceptor protein for the negative phototaxis of *E. halophila*. PYP has a photoreaction cycle comprised of several intermediates, the spectral and kinetic properties of which are very similar to those of the retinal proteins in halobacteria.

By flash photolysis at room temperature, After absorption of a blue photon, the ground state of PYP enters a photocycle in which a red-shifted intermediate, L($\lambda_{\text{Max}}=465\text{nm}$), and blue-shifted intermediate, M($\lambda_{\text{Max}}=355\text{nm}$), are formed sequentially, followed by the reformation of the ground state. In this photocycle PYP transduces a light signal into a protein conformational change that forms the physiological signal. M intermediate is expected to be the signaling state. M decays about 500 msec by flash photolysis at room temperature.

In this time, we measured native PYP by Small Angle X-ray Scattering (SAXS) at BL45XU. The radius of gyration (Rg) of native PYP was estimated by the Guinier plot of the

scattering profiles. The result of the experiment was that the Rg was 15.46 Å at pH 7.4. Although we have measured PYP by SAXS at PF in Tukuba, the result was that the Rg was 15.6 Å at pH 7.2. It seems reasonable to suppose that the result of this study corresponds with that of the data in PF. However the concentration dependence of Rg cannot be estimated by the Guinier plot of the scattering profiles. Though in different protein concentrations, 2, 4, 6, 8, 9, 10, 12 mg/ml, we measured, the Guinier plots of a concentration higher than 8 mg/ml are well-approximated by a straight line. So far, we have decided the measurement condition. In the future, by time-resolved SAXS the property of M intermediate of PYP, such as changing charge of molecule surface, will be revealed.