

Accessing protein conformations of green fluorescent protein NowGFP at cryogenic and room temperature using X-ray crystallography

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A green-emitting fluorescent protein variant, NowGFP, with a tryptophan-based chromophore (Thr65-Trp66-Gly67) was found to have the conformational changes induced at acidic pH, which are local active site rearrangement without disrupting the crystal packing. Here the X-ray structures of NowGFP were determined at pH 5.0 and pH 8.0 at cryogenic and room temperature using synchrotron and x-ray free electron laser sources. In a typical β -barrel structure, major conformational changes occur in Lys61 and hydrogen-bond networks comprising Lys61, Glu222, Thr203 and Ser205, which was connected to the indole ring of Trp66 of the chromophore at the active site. Based on structural differences at cryo and room temperature, time-resolved studies will be monitored by serial femtosecond crystallography.