Photoisomerization of bacteriophytochromes in crystals and in solutions - a time-resolved study

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Detection of atomic motions of reactions by means of free electron laser setups has become possible. An avenue is open for studies of reactions, such as isomerization processes, in proteins or in other complex (bio)materials, in atomic detail (1). Apart from the necessity for high-power X-ray pulses and advanced detection facilities, microcrystals which diffract to considerably high resolution (2.2 Å or lower) are needed. Here, we demonstrate by optical spectroscopic methods that the photochemical response of a bacteriophytochrome system is dependent on crystal packing, proper protonation state for photochemical reactions (2). At low pH (5.4), where the highest resolution crystals are formed, the photochemical yield of the first intermediate state (Lumi-R-state) is reduced ten-fold in comparison to the data observed at pH 8 in solution. Lumi-R production is completely abolished in the crystal state. Clearly, protonation, or proton-transfer changes are reflected in spectroscopic responses. Furthermore, dynamic constraints due to crystal contacts influence excited state dynamics, and thus Lumi-R formation. Consequently, we show that phytochrome protein scaffolds undergo considerable structural changes already in the early moments of the excited state.

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References

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