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Probing a Conformational Change of a Photoswitchable Allosteric Protein with Ultrafast IR Spectroscopy BRIGITTE STUCKI-BUCHLI, STEVEN A. WALDAUER, RETO WALSER, ROLF PFISTER, PETER HAMM, Department of Chemistry, University of Zurich, Switzerland — By covalently linking an azobenzene photoswitch across the binding groove of an allosteric protein domain, a conformational transition can be initiated by a laser pulse. This transition mimics the conformational change of the unmodified domain upon ligand binding. We have studied this light induced conformational change by ultrafast IR spectroscopy. So far, we have probed two IR absorption bands: First, the amide I band which arises from the carbonyl stretch vibration of all amide groups in the protein and is sensitive to overall structural changes, and second, a vibration localized on the photoswitch, which is sensitive to its local environment, namely the opening of the binding groove. We have found that the binding groove opens on a timescale of 100 ns in a non-exponential manner. Even after the binding groove has equilibrated, the protein conformation still continues to change elsewhere. Currently, we are incorporating site-specific IR labels, to learn more about the response of the protein to the perturbation of the binding groove.

¹Buchli, B. et al. Kinetic response of a photoperturbed allosteric protein. Proceedings of the National Academy of Sciences of the United States of America 110, 11725-30 (2013).

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