LABORATOIRE D'OPTIQUE ET BIOSCIENCES

Unité INSERM U1182 - UMR CNRS 7645 - Ecole Polytechnique

Ecole Polytechnique, 91128 Palaiseau cedex

Vendredi 10 décembre 2018 à 10h

Ecole Polytechnique

Amphithéâtre Curie

Steve Meech

University of East Anglia

Femtosecond to Microsecond Study of Mechanism in Photoswitchable Fluorescent Proteins

Photochromic fluorescent proteins play a critical role in super-resolution microscopy and have important applications in optogenetics. In particular the ability to control the on/off switching rate is critical in a number of applications, yet the detailed mechanism is yet to be determined. It is established that the light driven structure change which modulates the fluorescence involves both a trans to cis isomerization and a proton transfer. In this work we present a detailed experimental study of the 'off' to 'on' state switching mechanism in dronpa using femtosecond to millisecond time resolved infra-red spectroscopy coupled with isotope labelling. The labelling study shows that both chromophore and protein dynamics occur on multiple timescales, from picoseconds to hundreds of microseconds. Following excitation of the trans chromophore a ground state primary photochemical product is formed in picoseconds. Surprisingly the characteristic vibrational spectrum of the neutral cis isomer appears only after several tens of nanoseconds following a reorganisation in the electronic ground state. A further fluctuation in the protein structure around the neutral cis chromophore is required to form a new intermediate which acts as a gate to the final proton transfer reaction, leading ultimately to formation of the cis anion on-state in several tens of microseconds. These data illustrate the important interplay between changes in chromophore structure and protein environment underlying fluorescent protein photochromism.

Renseignements complémentaires

Marten VOS – Tel : 01 69 33 50 66 - e.mail: marten.vos@polytechnique.edu

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