## MS16 Time-resolved diffraction and scattering techniques

#### MS16-03

Steady-state and time-resolved X-ray scattering demonstrate that oligomerization processes limit photoactivation and recovery of the cyanobacterial Orange Carotenoid Protein

## E.A. Andreeva <sup>1</sup>, J.P. Colletier <sup>2</sup>

<sup>1</sup>Institut de Biologie Structurale (IBS), Univ. Grenoble Alpes, CEA, CNRS, 38044 Grenoble, France; Max-Planck-Institut für medizinische Forschung, Jahnstraße 29, 69120 Heidelberg, Germany - Grenoble (France), <sup>2</sup>Institut de Biologie Structurale (IBS), Univ. Grenoble Alpes, CEA, CNRS, 38044 Grenoble, France; - Grenoble (France)

#### Abstract

The photoactive Orange Carotenoid Protein (OCP), involved in cyanobacterial photo-protection [1], is a two-domain protein which upon light absorption transitions from a compact inactive "dark" state ( $OCP^{O}$ ) to an extended photoactive "light" state ( $OCP^{P}$ ) [2,3]. The molecular mechanism enabling photoactivation involves migration of the carotenoid pigment from the interface between the two domains into the NTD, and subsequent domain dissociation. It remains unclear what the exact structure of  $OCP^{R}$  is, on which timescale it forms, and whether or not oligomerization [2,4] is involved in the regulation of OCP function – either in the inactive or photo-activated state. Here, we used a combination of steady-state and time-resolved (TR) X-ray scattering and spectroscopy to address these three specific issues. Our results inform on the time scales associated to the dissociation and reassociation of domains in monomeric OCP, yielding  $OCP^{R}$  and enabling recovery of  $OCP^{O}$ , respectively. They furthermore demonstrate that oligomerization occurs at the  $OCP^{O}$  and  $OCP^{R}$  level, limiting the photoactivation of  $OCP^{O}$  and the  $OCP^{O}$  to  $OCP^{R}$  thermal recovery, respectively [5].

#### References

[1] Kirilovsky D. & Kerfeld C. A., (2016). Cyanobacterial photoprotection by the orange carotenoid protein. Nat. Plants 2, 16180.

[2] Wilson A. et al., (2008). A photoactive carotenoid protein acting as light intensity sensor. PNAS, 105 (33): 12075-12080

[3] Haijun L. et al., (2016). Dramatic Domain Rearrangements of the Cyanobacterial Orange Carotenoid Protein upon Photoactivation. Biochemistry,55 (7): 1003–1009

[4] Golub M. et al., (2019). Solution Structure and Conformational Flexibility in the Active State of the Orange Carotenoid Protein: Part I. Small-Angle Scattering. J. Phys. Chem. B, 123, 95259535

[5] Andreeva E.A. et al., (2022). Oligomerization processes limit photoactivation and recovery of the Orange Carotenoid Protein. Biophysical Journal (in review).



# TR X-ray scattering monitors OCP photoactivation

# Proposed model for OCP photoactivation

