Session: Fundamental Photochemistry & Photophysics

Effect of truncation of cytoplasmic tail on ultrafast photo-cycle of Anabaena Sensory Rhodopsin

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Anabaena Sensory Rhodopsin (ASR) is a microbial retinal protein (MRP) which converts the photon energy to chemical energy by a cis-trans isomerization. This energy is stored as vibrational motion of retinal chromophore and utilized latter to expel protons from cytoplasmic region to outside the cell. ASR is a unique member in rhodopsin family which can bind both all-Trans (AT) and 13-Cis (13C) isomers in same protein environment but the isomeric ratio changes depending on chromatic adaptation condition.[1] In dark-adapted (DA) state, more than 95% of chromophores adopts AT conformation whereas in light-adapted (LA) condition, it binds both AT and 13C isomer. 13C isomer shows blue shifted ground state absorption (fig.1(a)) . Also, it shows faster dynamics and smaller quantum yield compared to AT form.[2] In the past, it has been observed that electrostatic interaction of opsin with the retinal chro-

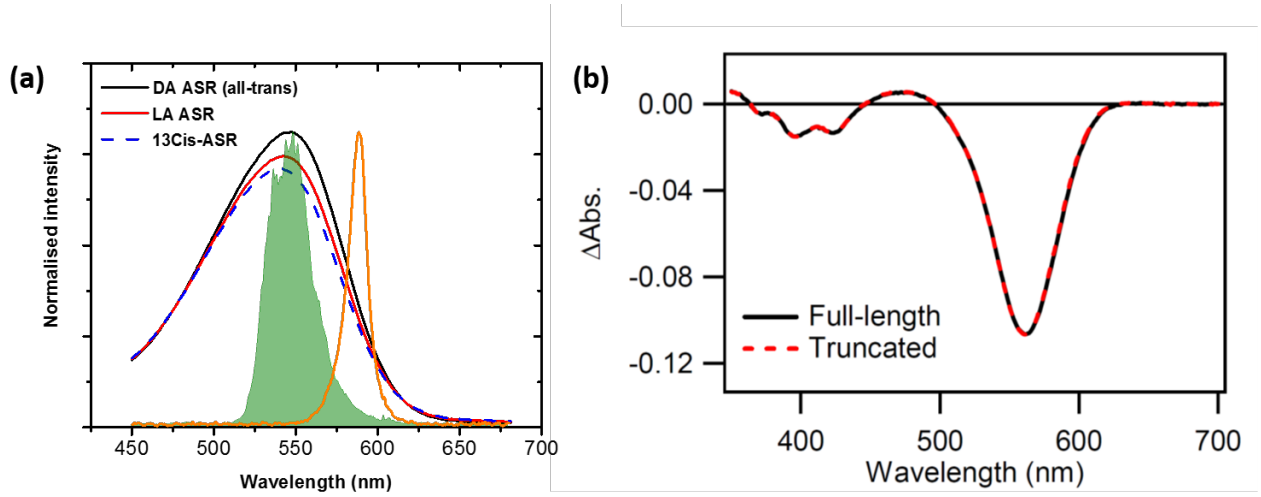

Figure 1: (a) Ground state absorption of ASR in Dark and Light adapted state and extracted pure 13C isomer spectra. Orange and green curve represent illumination spectra for light adaptation and excitation spectra for pump-probe study (c) Light minus dark difference absorption spectra for Full length and Truncated ASR at 25° C.

mophore determines the ultrafast dynamics.[3] Slight change in environment, by changing pH or solvent or by point mutation, can alter the dynamics. It has been reported[4] that truncation of cytoplasmic domain (after 229 residues) of ASR inverts the direction and rate of proton

movement during Schiff base deprotonation. On the other hand, truncation has been found be advantageous for higher level of expression and also for crystallization.[5]

In this report, we used femtosecond transient absorption (TA) spcectroscopy to compare the sub-10 ps dynamics of Full length (FL) and Truncated (Tru) form of ASR to explore the importance of cytoplasmic tail on primary photo-cycle of ASR. The excitation pulse was resonant (545 nm) to the steady state absorption of the chromophore (fig.1(a)). The broadband visible white light, covering 410-700 nm, was utilized for probing. The global analysis parameters of TA spectra of FL ASR agrees well with the previous study.[2] Our study shows that truncation of the C-domain does not affect the isomeric ratio and ground state absorption spectra (fig.1(b)) but there are three major differences between FL and Tru ASR: (i) the rise of photo-product is slightly faster (τ=800 fs) for Tru form than FL form (τ=710 fs); (ii) in LA condition, Tru form

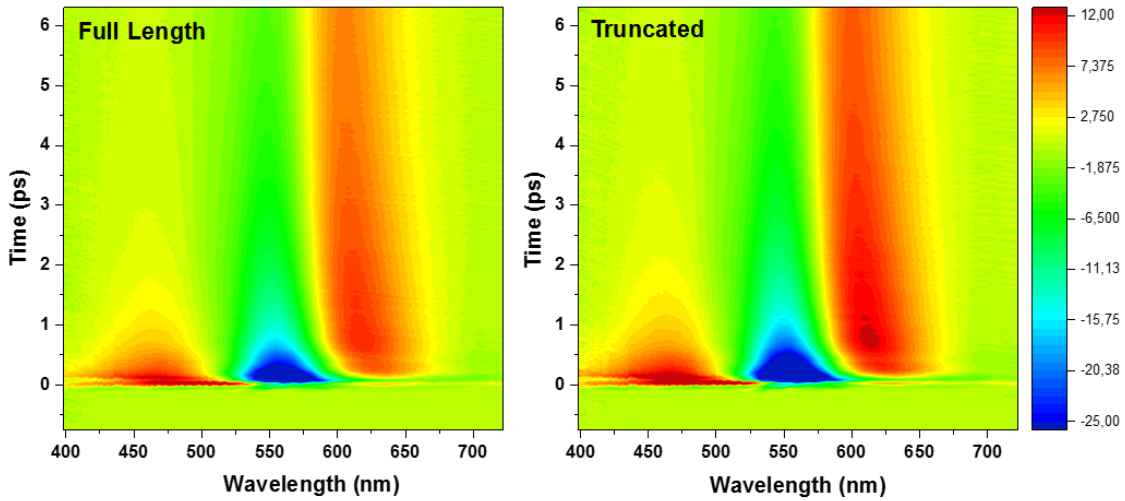

Figure 2: Spectro-temporal evolution of Transient absorption difference spectra (ΔOD) for Full length and Truncated ASR in light adapted condition.

shows higher amplitude (fig.2) in TA signal around 620 nm which is known to be the region of photo-product (hot 'J' product) absorption; (iii) DA kinetics is found to be slower (τ= 152 vs 83 min) for Tru ASR.

Funding: Deutsche Forschungsgemeinschaft (DFG)

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