Structural differences between the closed and open states of channelrhodopsin-2 as observed by EPR spectroscopy

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Most ion channels are triggered by binding ligands or are voltage gated, whereas channelrhodopsin-2 (ChR2), a phototaxis receptor of algae, is a cation channel activated by light [1]. This unique property is employed to optically trigger action potentials and other cellular activities [2]. Besides its broad application in neurological research, light excitability makes ChR2 accessible to a broad range of spectroscopic techniques even on ultrafast timescales with the aim to address the functional mechanism of this unique membrane protein. The recently published structure on a ChR1/ChR2 hybrid resolved at a resolution of 2.3 Å [3] provides a firm basis for probing structural changes at different positions during channel activity.
Two variants which contain either 1 or 2 wild-type cysteines were derivatised with nitroxide spin label and subjected to pulsed electron double resonance (pELDOR) spectroscopy. Both variants contained the C128T mutation [4] to trap the long-lived P3520 state by illumination as demonstrated by FTIR difference spectroscopy. Comparison of spin-spin distances in the dark state and after illumination reflect conformational changes in the conductive P3520 state involving helices B and F (figure 1). Spin distance measurements reveal that ChR2 forms a dimer in the absence of intermolecular N-terminal cysteine.

References:

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figure 1