Photocycle of Bacteriorhodopsin in Lipid Nanodisc

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Abstract

Incorporating membrane proteins into membrane mimicking systems has been a tricky process for subsequent biophysical characterizations. Monodisperse lipid nanodisc, a high density lipoprotein composed of a lipid bilayer wrapped with two copies of membrane scaffold protein, is a popular platform for studying membrane protein in near-native environment. By incorporating bacteriorhodopsin (bR) in nanodiscs composed of different lipid compositions, we investigated the effect of lipid environment on the function of the membrane protein. The structural conservation of bR in nanodisc was shown in steady-state absorption contours, while the changes of the lifetime of intermediate states of bR in nanodiscs were revealed using full-wavelength transient absorption spectroscopy. Our results indicate that one can manually tune the photocycle kinetics of bR using nanodiscs with different lipid composition. To further improve the native environment, we show a one-step incorporation of trimeric bR into lipid nanodisc directly from the native purple membrane of *Halobacterium Salinarum,* without addition of any synthetic lipids or lipid extracts. The method was demonstrated to produce sufficiently high yield suitable for biophysical studies. Circular dichroism spectra were collected to show the trimeric conformation of bR was maintained, and the transient absorption spectra showed that the photocycle activity of bR incorporated in native membrane nanodisc was preserved. Our results suggest that the surrounding lipids can alter the biological functions of membrane protein, and that it is possible to transfer membrane protein directly from native membrane to lipid nanodisc.