Structural Changes of Cephalopod Rhodopsin and β-Arrestin Measured by FTIR Difference Spectroscopy and Isotope Editing

Joel M Kralj1, Erica Raber1, Jose Sarmiento2, David Shumate2, Christie Stanzel2,
Carrie Maxwell2, Javier Navarro2 and Kenneth J Rothschild1

1Department of Physics, Molecular Biophysics Laboratory, Photonics Center, Boston University, Boston, Massachusetts 02215, USA
2Department of Neuroscience and Cell Biology, University of Texas Medical Branch, Galveston, Texas 77555, USA

Abstract:
Invertebrate rhodopsin is the primary photoreceptor found in the eyes of cephalopods. Unlike vertebrate rhodopsin, invertebrate rhodopsins such as sepia rhodopsin (s-Rh) can be activated by light and then rapidly cycled back to the original state with a second red-shifted photon, thereby facilitating a variety of novel biophysical studies. Additionally, invertebrate rhodopsins can bind to the ubiquitous β-Arrestin which is involved in regulating signal transduction in many GPCRs. In this study, we used static and time-resolved FTIR difference spectroscopy to investigate the photocycle of s-Rh complexed to β-Arrestin. In the spectrum of s-Rh alone, difference spectra obtained using two colors to cycle between the ground state (cis) and acid meta state show an 11- to all-trans behavior in the presence of arrestin as evidenced by the rate of photobleaching.

Conclusions:
Sepia rhodopsin shows altered behavior in the presence of arrestin as evidenced by the rate of photobleaching. The presence of arrestin can be detected in the FTIR difference spectrum mainly in the amide I and II regions which are sensitive to protein backbone vibrations. Labeling just the arrestin with 15N total isotope results in a shift of difference bands indicating that the arrestin is undergoing a conformational change during the rhodopsin two photon cycle. The frequencies of the arrestin bands that change indicate it reflects alpha-helical structure undergoing changes, consistent with the alpha-helix anchor model.

References:

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