Structure of Rhodopsin as a G Protein-Coupled Receptor

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In rhodopsin, light-induced isomerization of the 11-*cis*-retinal to its all-*trans* form initiates conformational changes within the transmembrane protein that enable the activated rhodopsin to bind and activate the heterotrimeric G protein transducin on the cytoplasmic surface. The structure of rhodopsin (PDB code 1GZM) was recently determined at 2.65Å resolution [1, 2] in a $P3_1$ crystal form where the packing retains an amphipathic molecular environment similar to that in a membrane, so the transducin-interaction segments from the cytoplasmic interface are likely to represent their native conformations. By docking this crystal structure into a cryo-EM map of 2D crystals [3] we consider the mechanistically important details in the membrane context.

The kinked transmembrane helices enfold the retinal in a tight binding pocket. In the membrane interior, ordered water molecules mediate key interactions. Glu113 with a water molecule hydrogen bonded between its main chain and side chain oxygen atoms forms a complex counterion for the protonated Schiff base linking the retinal to helix H7, which may then stabilise the salt bridge with the protonated Schiff and help to stabilise the dark state. Other water molecules extend the inter-helical hydrogen bonding networks, linking Trp265 in the retinal (ligand) binding pocket to the NPxxY motif on the cytoplasmic side, important for G protein-coupling, and the Glu113 counterion to the extracellular surface. Modulation of these networks is likely to be involved in the activation process.

The G protein interaction sites mapped to the cytoplasmic ends of H5 and H6 and a spiral extension of H5 are elevated above the bilayer. The conserved Glu134-Arg135 ion pair in H3 is sequestered in the walls of a surface cavity. The highest temperature factors in the cytoplasmic loops suggest that they are quite flexible when not interacting with G protein or regulatory proteins. A tightly bound detergent molecule wraps around the kink in H6, stabilizing the structure around the potential hinge in H6. These findings support an activation mechanism that involves pivoting movements of kinked helices, which, while maintaining hydrophobic contacts in the membrane interior, can be coupled to amplified translation of the helix ends near the membrane surfaces.

[1] Li J., Edwards P.C., Burghammer M., Villa C., Schertler G.F.X., *J. Mol. Biol.*, 2004, **343**, 1409. [2] Edwards P.C., Li J., Burghammer M., McDowell J.H., Claudio V., Hargrave P.A., Schertler G.F.X., *J. Mol. Biol.*, 2004, **343**, 1439. [3] Krebs A., Edwards P.C., Villa C., Li J., Schertler G.F.X., *J. Biol. Chem.*, 2003, **278**, 50217-50225.

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