Structure and gating mechanism of the nicotinic acetylcholine receptor

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The nicotinic acetylcholine (ACh) receptor is the neurotransmitter-gated ion channel at the nerve-muscle synapse. It serves as a model for other members of the Cys-loop superfamily, including neuronal ACh, $GABA_A$, glycine and $5\text{-}HT_3$ receptors. Structures of the ACh receptor in the closed-channel form (Unwin, 2005; Miyazawa, Fujiyoshi & Unwin, 2003) and in the open-channel form (Unwin, 1995) are now known to resolutions of 4Å and 9Å respectively, from electron crystallographic studies of *Torpedo* postsynaptic membranes.

The receptor is a large (290kD) glyco-protein, assembled from a ring of homologous subunits (α, γ, α, β, δ) and divided into three domains: a large N-terminal extracellular ligand-binding domain, a membrane-spanning pore, and a smaller intracellular domain, giving it a total length of about 160Å normal to the membrane plane. The ligand-binding domain shapes a long, ~20Å diameter central vestibule and has two binding sites for ACh, which are in the α subunits at interfaces with the γ and δ subunits, on opposite sides of the pore. The pore makes a narrow water-filled path across the membrane and contains a hydrophobic gate, which breaks open when ACh occupies both binding sites. The intracellular domain shapes another, smaller vestibule, having narrow lateral openings for the ions. The inner surfaces of both vestibules are negatively charged, creating a cation-stabilising environment at either entrance to the membrane pore.

We show that the ligand-binding portions of the two α subunits have a ‘distorted’ conformation relative to the β, γ and δ subunits when the binding sites are empty and the channel is closed. Binding of ACh causes a local rearrangement in which loops B and C of the α subunits are drawn in around the bound ligand to enable coordination of relevant side-chains. The local rearrangement overcomes the distortions in the α subunits, stabilising an alternative conformation which is like that of the non-α subunits. This transition is accompanied by rotations of the inner sheet of the β sandwich composing the ligand-binding portions of the α subunits. The rotations are communicated to the pore-lining α-helices in the membrane, triggering cooperative movements which disrupt the gate of the channel, allowing ions to flow through. Thus gating in this channel appears to occur by fast cooperative movements of helices lining the membrane pore, whereas the ligand-binding domain appears to function as a controlling device that either disenables or facilitates these movements.

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Miyazawa, A., Fujiyoshi, Y. & Unwin, N. (2003) *Nature* **423**, 949-955.
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