s1.m7.o2 **Crystal Structure of Oxido Squalene Cyclase.**
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Keywords: Membrane Protein; Drug Design; Cyclase

Oxido Squalene Cyclase (OSC) catalyses the synthesis of the steroid scaffold which is a key step in the synthesis of cholesterol. In a single highly selective reaction OSC forms lanosterol from the linear substrate oxido squalene.

The 2.1Å structure of this human monotopic integral membrane protein shows how OSC is inserted to the membrane. The hydrophobic substrate can reach the active site that is deeply buried in the center of the enzyme through a channel that opens into the membrane. The structure gives also new insights into the way OSC catalyzes the interesting cyclization reaction. Analysis of the mode of inhibitor binding to the active site cavity will help in the design of new OSC inhibitors as anticholesteremic drugs.

Also the high level expression, purification and crystallization of this human membrane protein will be described. Analytical ultra centrifugation was used to characterize the aggregation state of OSC and was helpful in predicting crystallizability.

s1.m7.o3 **Activation of a Vinculin Binding Site (VBS) in the Talin Rod Involves Rearrangement of a Five Helix Bundle.**
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Keywords: Talin; Vinculin; Integrin

The integrin family of adhesion receptors provide multicellular organisms with a physical link between the extracellular matrix (ECM) and the actin cytoskeleton. Integrins form intracellular signalling complexes called focal adhesions. Talin is a highly conserved cytosolic protein that plays a key role is focal adhesion assembly and forms a vital link between integrins and the actin cytoskeleton Talin functions by binding directly to the short cytoplasmic domains of integrin b subunits, to F-actin and several other focal adhesion component proteins including vinculin. Talin has a 50kDa N-terminal head domain and a FERM domain (residues 86-400). The remainder of the protein is comprised of a 220kDa rod domain which is dimeric, contains three binding sites for the cytoskeletal protein vinculin [1], [2]. We have determined the first crystal structures of two domains from the rod region of the talin spanning residues 482-789. The talin 482-789 structure is composed of an N-terminal 5-helix bundle and a C-terminal 4-helix bundle connected by a short loop. The two domains form a flattened elongated Z shape structure with dimensions 90Å×45Å×25Å. Within the 5-helix bundle (talin 482-655), helices H2-H5 fold into a right handed up and down 4-helix bundle. Helix H2 is elongated compared to the other helices and bends allowing a 12 residue loop to reach over H3 and connect to H1. The addition of H1 to the H2-H5 fold represents a novel topology, and results in the structure having a triangular rather than square equatorial cross section. Talin (482-655) contains a vinculin binding site (VBS). We show that the VBS is composed of a hydrophobic surface spanning five turns of an α-helix. All the key sidechains from the VBS are buried and contribute to the hydrophobic core of the talin 482-655 fold. We demonstrate the talin 482-655 5-helix bundle represents an inactive conformation; mutations which disrupt the hydrophobic core or deletion of helix 5 are required to induce an active conformation in which the VBS is exposed. Activation of the VBS in talin, and the recruitment of vinculin may support the maturation of small integrin/talin complexes into more stable adhesions. We have also determined the crystal structure of talin (482-636) in complex with the Vinculin head domain. This reveals the mechanism underlying the interaction between the two proteins as well as the resulting conformational changes.

[1] Critchley, D.R. (2000) *Curr Op Cell Biol.***12**, 133-139
[2] Garcia Alvarez *et al.*, (2003) *Mol. Cell.* **11**, 49-58