THE BASAL ACTIVITY OF CONSTITUTIVE ANDROSTANE RECEPTOR (CAR) - HOMOLOGY MODELLING VERSUS X-RAY CRYSTALLOGRAPHY

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The constitutive androstane receptor (CAR, NR1I3) belongs to the superfamily of nuclear hormone receptors that function as ligand-activated transcription factors. CAR plays an essential role in the metabolism of hormones and xenobiotics. In contrast to related nuclear hormone receptors CAR exhibits a constitutive activity for which the structural basis remains unclear.

To investigate the basal activity of CAR and the effect of co-activator binding homology models of the ligand binding domain (LBD) were generated [1]. Available x-ray structures of the related pregnane X (PXR) and the vitamin D receptor (VDR) were used as templates. Molecular dynamics (MD) simulations of CAR alone and in complex with a co-activator peptide (SRC-1) revealed a hypothesis for the activation mechanism. The basal activity of CAR can be explained by specific interactions between amino acids on the LBD and its C-terminal activation domain (AF-2).

To support the derived activation hypothesis, site directed mutagenesis studies were carried out on amino acids of the ligand binding pocket (LBP) [2]. In addition virtual receptor mutants were created and examined by MD simulations. The results not only support the proposed mechanism of constitutive activity but also give insights into the structural changes in the ligand binding pocket (LBP) upon mutation.

Docking studies carried out with program GOLD yielded the interaction modes of structurally diverse agonists unravelling the mechanisms by which ligands enhance CAR activity.

Compared to recently published x-ray structures of human CAR (1XV9, 1XVP) our homology model shows only minor deviations. However, the crystal structures - complexed with agonists - contain an unique additional helix that is supposed to keep the receptor in an activated state. It remains unclear whether this newly found helix is involved in maintaining constitutive activity or rather an effect of agonist binding, indeed.

[1] Windshügel et al. (2005) *J Mol Mod*, published online

[2] Jyrkkärinne et al. (2005) *J Biol Chem*, published online