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Transcriptional Regulation by the Nuclear Receptors Steroidogenic Factor-1 and Liver Receptor Homologue-1

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ABSTRACT

Steroidogenic Factor-1 (SF-1) and Liver Receptor Homologue-1 (LRH-1) are transcription factors belonging to the nuclear receptor (NR) family. In the adrenal cortex and gonads, SF-1 governs the expression of multiple enzymes and transporters required for converting cholesterol to steroid hormones. LRH-1 is mainly expressed in the enterohepatic system and regulates transcriptions of enzymes and transporters important in the conversion of cholesterol to bile acids. While most NRs bind DNA as either homo- or heterodimers, both SF-1 and LRH-1 interact with their response elements as monomers. On target gene promoters, they recruit coregulators that can either activate or repress transcription. Work presented in this thesis aims at elucidating the molecular mechanisms of SF-1 and LRH-1 actions in transcriptional regulation.

In Paper I we describe how a previously unknown modulator of SF-1 activity, RNF31, is important in DAX-1-dependent repression of SF-1. RNF31 is an E3 ubiquitin ligase and we show that it can monoubiquitinate DAX-1. RNAi-mediated knockdown of RNF31 and presumed loss of DAX-1 ubiquitination leads to increased expression of the SF-1 target genes StAR and aromatase (CYP19). We also show that RNF31 is present on the promoters of StAR and aromatase together with DAX-1 and SMRT and that its presence is SF-1-dependent. In conclusion, RNF31 is shown to be a novel coregulator of SF-1, acting via DAX-1 to repress transcription of StAR and aromatase.

In Paper II we further explore the role of RNF31 and SF-1 in adrenocortical cells. We knock down either RNF31 or SF-1 using RNAi, and use microarray analysis to identify differentially expressed genes. The cells depleted of RNF31 are shown to have significant changes in pathways related to steroidogenesis and cholesterol metabolism and many known SF-1 target genes are among those altered by RNF31 loss. This adds further support to our hypothesis that RNF31 is a coregulator of SF-1 activity. SF-1 depletion alters, besides steroidogenesis, pathways governing cell proliferation and differentiation. Genes involved in TGFβ- and Wnt/β-catenin-signalling are upregulated in response to SF-1 knockdown indicating that SF-1 signalling has repressive effects on these pathways.

In Paper III we show that LRH-1 is involved in transrepression of the hepatic acute phase response. The repression is dependent on agonist activation of LRH-1 and occurs in only a subset of the acute phase proteins. The regulated proteins have in common that the NCoR/HDAC3/GPS2-corepressor complex is recruited to the promoters. Upon ligand-activation, SUMOylated LRH-1 is tethered to and stabilises the corepressor complex, hindering its dissociation from the promoter and thus transcription of the target gene.

In conclusion, the major findings presented in this thesis are the characterisation of RNF31 as a coregulator of SF-1 and steroidogenesis, of GPS2 as a coregulator of SUMOylation-dependent transrepression by LRH-1 and the new, active role of ubiquitin-like modifications in transcriptional regulation by both LRH-1 and SF-1.

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