ACTIVATED STEROL REGULATORY ELEMENT-BINDING PROTEIN-2 (SREBP-2) SUPPRESSES HEPATOCYTE NUCLEAR FACTOR-4-MEDIATED CYP3A11 GENE EXPRESSION IN MOUSE LIVER

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[Purpose] SREBP-2 is a key transcription factor for the cholesterol homeostasis. Recent studies have suggested the association of CYP3A, major drug-metabolizing enzymes, with cholesterol metabolism. We previously found that SREBP-2 inhibits HNF-4α-mediated Cyp3a11 expression in reporter gene assays. However, the mechanism remains unknown. In the present study, we have investigated a mechanism for the SREBP-2-mediated inhibition of hepatic Cyp3a11 expression.

[Methods] Pull-down assays were performed with GST-fused HNF-4α or SREBP-2, and in vitro synthesized SREBP-2 and PGC-1α. Reporter assays were performed with Cyp3a11 reporter plasmids containing 5' flanking regions of Cyp3a11, and SREBP-2 and PGC-1α expression plasmid. Chromatin immunoprecipitation (ChIP) assays were performed with mouse livers.

[Results and Discussion] SREBP-2 showed the interaction with HNF-4α in pull-down assays, but was unable to interact with HNF-4α bound to DNA in the assays. Furthermore, SREBP-2 inhibited the interaction of HNF-4α with PGC-1α through the binding to PGC-1α. PGC-1α overexpression relieved the SREBP-2-mediated reduction of Cyp3a11 expression in reporter assays. ChIP assays demonstrated that the extent of PGC-1α binding to the Cyp3a11 promoter was reduced in mouse livers by feeding a low-cholesterol diet, which activates hepatic SREBP-2.

[Conclusions] Activated SREBP-2 is suggested to interact with PGC-1α in mouse livers at a condition of reduced cholesterol intake. This may result in the reduced PGC-1α recruitment to HNF-4α on the Cyp3a11 promoter and the subsequent down-regulation of Cyp3a11 expression.

PREGNANE X RECEPTOR MEDIATED INDUCTION OF MDR1A IS DIFFERENT FROM CYP3A11 IN TISSUE-, REGION- AND TIME-DEPENDENCIES IN MICE

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[Purpose] Expressions of P-glycoprotein (MDR1) and cytochrome P450 3A (CYP3A) are regulated by pregnane X receptor (PXR) in a ligand-dependent manner. However, the tissue-, region- and time-dependent regulation of these genes in vivo remains poorly understood. The purpose of this study was to examine the effects of pregnenolone 16α-carbonitrile (PCN), a potent ligand of murine PXR, on the induction of Mdr1a and CYP3A11 in the liver, intestine and brain in mice.

[Methods] Male C57BL/6 mice were treated with PCN once daily for 4 days. Tissues were collected at 8 or 24 hours after the final administration, and levels of Mdr1a, CYP3A11 and PXR mRNAs were determined by real-time PCR.

[Results and Discussion] In the liver, both Mdr1a and CYP3A11 were induced by PCN treatment. However, the extents of Mdr1a induction but not CYP3A11 were dependent on a time from the final treatment with PCN. In the intestine, the extent of Mdr1a induction was comparable in every section from duodenum to colon. In contrast, CYP3A11 was most inducible in the duodenum, and the extent of induction gradually decreased from duodenum to colon. In the whole brain, neither Mdr1a nor CYP3A11 were induced by PCN treatment. However, mRNA of Mdr1a but not CYP3A11 was increased by PCN treatment in the cortex, suggesting that induction of Mdr1a may occur at specific region of the brain.

[Conclusions] There were differences in tissue-, region- and time-dependency of Mdr1a and CYP3A11 induction by PCN, suggesting that different mechanisms are involved in the PXR-mediated induction of these genes in mice in vivo.