DOWN-REGULATION OF RENAL ORGANIC ANION TRANSPORTERS rOAT1 AND rOAT3 BY ISCHEMIA/REPERFUSION OF RAT KIDNEY
Takanobu Matsuzaki1, Kanako Yoshitome1, Takaumi Morisaki1, Hiroshi Watanabe1, Akinobu Hamada1, Yukimasa Kohda2, Hiroshi Nonoguchi2, Kimio Tomita2 and Hideyuki Saito1
1Department of Pharmacy, Kumamoto University Hospital, 1-1-1 Honjo, Kumamoto 860-8556 and 2Department of Nephrology, Kumamoto University Graduate School of Medical Sciences, 1-1-1 Honjo, Kumamoto 860-85596, Japan

The effect of acute renal failure (ARF) induced by ischemia/reperfusion (I/R) of rat kidney on the expression of organic anion transporters (OATs) was examined. The level of serum indoxyl sulfate (IS), a uremic toxin and substrate of OATs in renal tubules, shows a marked increase with the progression of ARF. However, this increase was significantly attenuated by ingestion of cobalt. The level of mRNA and protein of both rOAT1 and rOAT3 were markedly depressed in the ischemic kidney. The uptake of p-aminohippuric acid (PAH) and estrone sulfate (ES) by renal slices of ischemic rats was significantly reduced compared to control rats. Renal slices taken from ischemic rats treated with cobalt displayed significantly elevated levels of ES uptake. Cobalt intake did not affect PAH uptake, indicating the functional restoration of rOAT3 but not rOAT1. The expression of Na+/K+-ATPase was markedly depressed in the ischemic kidney, suggesting that the inward Na+ gradient in renal tubular cells had collapsed, thereby reducing the outward gradient of α-ketoglutarate, a driving force of both rOATs. The decreased expression of Na+/K+-ATPase was significantly restored by cobalt treatment. Our results suggest that the down-regulation of renal rOAT1 and rOAT3 could be responsible for the increase in serum IS level of ischemic rats. Cobalt treatment has a significant protective effect on ischemia-induced ARF, being accompanied by the restoration of rOAT3 and/or Na+/K+-ATPase function.

CHOLESTEROL-DEPENDENT EXPRESSION OF NIEMANN-PICK C1 LIKE 1
Yuki Iwayanagi, Tappei Takada and Hiroshi Suzuki
Department of Pharmacy, The University of Tokyo Hospital, Faculty of Medicine, The University of Tokyo, Tokyo 113-8655, Japan

Niemann-Pick C1 like 1 (NPC1L1), which is recently identified as a target of ezetimibe, a novel cholesterol lowering drug, is expressed mainly in the liver and small intestine and is believed to be important for the distribution of cholesterol in the body. Some of the genes involved in cholesterol homeostasis, such as low density lipoprotein receptor and 3-hydroxy-3-methylglutaryl-CoA (HMG-CoA) reductase, are known to be negatively regulated by sterols via transcriptional factors, sterol responsive element binding proteins (SREBPs). Considering the involvement of NPC1L1 in the uptake of sterols into cells, we hypothesized that NPC1L1 is also a target of SREBPs and its expression is dependent on cellular cholesterol level. The results of in vivo studies that the expression of NPC1L1 mRNA in mouse liver was decreased by cholesterol feeding are consistent with this hypothesis. Endogenous expression of NPC1L1 in HepG2 cells was also cholesterol-dependent. Cells treated with sterols had lower expression of NPC1L1, whereas HMG-CoA reductase lovastatin treatment resulted in the enhanced expression of NPC1L1, due to reduced cellular cholesterol levels. Reporter gene assay using 5'-flanking region of NPC1L1 showed that co-transfection of active SREBP2 with reporter vectors into HepG2 cells increased the transcriptional activity. Additionally, SREBP-dependent transactivation of NPC1L1 was further stimulated by hepatocyte nuclear factor-4alpha (HNF-4alpha). Involvement of putative SREs and HNF-4 binding elements in the promoter region of NPC1L1 is to be clarified in further investigation.