GENE REGULATION OF DRUG TRANSPORTERS
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Drug transporters play critical roles for the absorption, distribution and excretion of drugs, and have been classified into five major families, ABC transporter, peptide transporter (PEPT), organic anion transporting polypeptide (OATP), organic ion transporter (OAT/OCT/OCTN) and H+/organic cation antipporter (MATE). The latter four families were categorized as solute carrier (SLC) members, and molecular information such as structure, function and expression have been accumulated. Various studies also demonstrated the regulatory aspects of SLC drug transporters in the physiological and pathophysiological conditions (1), but molecular mechanisms underlying the gene regulation have not been fully understood. In addition to biochemical signals, genetic and epigenetic factors also affect the expression of SLC drug transporters. In this symposium, we will talk about our recent studies on the regulation of drug transporters.

Starvation markedly increased the amount of mRNA and protein of intestinal PEPT1, and the fasting-induced expression of PEPT1 altered the pharmacokinetic profiles of β-lactam antibiotic (2). Several lines of in vivo and in vitro studies have revealed that nuclear receptor, peroxisome proliferator-activated receptor α (PPARα), contributes to the augmentation of PEPT1 during fasting (3). The intestinal PEPT1 also showed the diurnal rhythm with a peak near the onset of darkness in rats feeding ad libitum (4), and that this diurnal rhythm was inverted by daytime (9:00 ~ 15:00) restricted feeding (5). We compared the daily fluctuations in the intestinal PEPT1 mRNA expression with those of several transcription factors, Sp1 (basal transcription factor), Cdx2 (intestine-specific transcription factor), PPARα and clock genes in rats between feeding ad libitum and daytime restricted feeding. Among these transcription factors, the expression of DBP (clock-controlled gene) showed similar phase with that of PEPT1 in both groups. Electrophoretic mobility shift assay (EMSA) and reporter assay revealed that DBP has the ability to bind the DBP-binding-site located in the distal promoter region of the rat PEPT1 gene and induce the transcriptional activity. These findings indicate that DBP contributes to the circadian oscillation of PEPT1 (6).

The core promoter and proximal promoter regions contain the elements which control the initiation of transcription, and therefore, essential regions that carry functionally relevant polymorphisms may have significant effects on gene expression. Based on these ideas, we searched single nucleotide polymorphisms (SNPs) in the promoter region of organic ion transporters and MATE1, and found several polymorphisms (7-8). For example, a SNP in the MATE1 gene (-32G>A) which belongs to a Sp1-binding site was identified, and we demonstrated that this SNP induced the decrease of Sp1-binding and promoter activity (8). Upstream stimulating factor (USF) 1 functions as a basal transcriptional regulator of human OCT2 gene via the E-box (9). USF1 is ubiquitously expressed, whereas human OCT2 is predominantly expressed in the kidney. Because epigenetic regulation such as DNA-methylation has been demonstrated to be involved in the tissue specific expression of several genes, we examined the effect of DNA methylation on the kidney specific expression of OCT2. All CpG sites in the OCT2 proximal promoter were hypermethylated in the liver, while hypomethylated in the kidney. The level of methylation of the OCT2 promoter was especially low at the CpG site in the E-box. In vitro methylation of the OCT2 proximal promoter dramatically reduced the transcriptional activity and EMSA showed that methylation at the E-box inhibited the binding of USF1. These results indicate that the kidney-specific expression of human OCT2 is regulated by methylation of the proximal promoter region, interfering with the transcription by USF1 (10).

References: