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Regulation of mRNA Expression of Xenobiotic Transporters by the Pregnane-X Receptor (PXR) in Mouse Liver, Kidney, and Intestine

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List of nonstandard abbreviations: PCN, pregnenolone-16 α -carbonitrile; Cyps, cytochrome p450 enzymes; Oatp, organic anion transporting polypeptide; Mdr, multidrug resistance protein; Mrp, multidrug resistance-associated protein;

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Abca1, ATP-binding cassette (Abc) transporter subfamily a1; Cnt, concentrative nucleoside transporter; Ibat, ileal bile acid transporter; Bsep, Bile salt export pump; Bcrp, breast cancer resistance protein; bDNA assay, branched DNA signal amplification assay; Npc1L1, Niemann-pick C1 like 1 transporter.

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Abstract

Multiple transporter systems are involved in the disposition of xenobiotics and endogenous compounds. The pregnane-X receptor (PXR) is a major chemical sensor known to activate the expression of CYP3A/Cyp3a in humans and rodents. The purpose of this study is to systematically determine whether the major xenobiotic transporters in liver, kidney, duodenum, jejunum, and ileum are induced by pregnenolone-16 α -carbonitrile (PCN), and whether this increase is mediated by the nuclear receptor PXR. In liver, PCN induced the expression of Oatp1a4 and Mrp3 mRNA in wild-type (WT) mouse liver, but not in PXR-null mice. In kidney, PCN did not alter the expression of any drug transporter. In duodenum, PCN increased Abca1 and Mdr1a mRNA expression in WT mice, but not in PXR-null mice. In jejunum and ileum, PCN increased Mdr1a and Mrp2 mRNA, but decreased Cnt2 mRNA in WT mice, but none of these transporters was altered when PCN was administered to PXR-null mice. Therefore, PCN regulates the expression of some transporters, namely Oatp1a4 and Mrp3 in liver, as well as Abca1, Cnt2, Mdr1a, and Mrp2 in small intestine via a PXR-mediated mechanism.

Introduction

The pregnane-X receptor (PXR, NR1I2), also called SXR (steroid-xenobiotic receptor) or PAR (pregnane-activated receptor), is an orphan nuclear receptor first identified by Kliewer *et al* (Lehmann *et al.*, 1998). PXRs have been characterized in rats, mice, humans, rabbits, and chickens. Mouse PXR shares 95% sequence similarity to rat, and 77% to human PXR (Zhang *et al.*, 1999; Moore *et al.*, 2003).

PXR is a master regulator of phase-I and -II metabolism of xenobiotics. Upon ligand binding, PXR forms a heterodimer with the retinoid-X receptor alpha (RXR α) and then transactivates target genes. PXR ligands generally induce Cyp3a/CYP3A genes in rats, mice, and humans (Goodwin *et al.*, 2002; Guo *et al.*, 2002b; Cheng *et al.*, 2005b). One exception is CYP3A41, which is down-regulated by DEX in a PXR-dependent manner (Anakk *et al.*, 2004). PCN also regulates phase-II drug metabolizing enzymes, such as glutathione-S-transferase (Gst) α 1 and μ 1, rat UDP-glucuronosyltransferase 1a1 (Ugt1a1) (Chen *et al.*, 2003), and sulfotransferase (Sult) 1b1 (Dunn *et al.*, 1999). In addition, PCN induces the expression of some transporters, such as rat Abca1 (Sporstol *et al.*, 2005), human MRP2, and MRP3 (Kretschmer and Baldwin, 2005).

The regulation of hepatic and small intestinal gene expression by two potent rat PXR agonists, L-742694 and DEX, has been examined after oral administration to female Sprague-Dawley rats using DNA microarray analysis (Hartley *et al.*, 2004). Both L-742694 and DEX elicited robust increases in gene expression of Cyp3a1/23, Cyp3a18, Ugt1a1, Ugt2b1, and Oatp1a4 in rat liver.

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However, both L-742694 and DEX suppressed rat Cyp3a1/23, Cyp3a18, Ugt1a1, Ugt2b1, Oatp1a4, and Mrp3 expression in intestine (Hartley et al., 2004). There was a distinct lack of coordinated inter-organ expression of genes common to both hepatic and intestinal tissues in rats (Hartley et al., 2004).

Mice and rats are two common species used to study regulatory mechanisms of drug metabolizing enzymes. PCN, a synthetic steroid, is a potent rodent PXR activator. The purpose of this study was to systematically investigate the regulation of major drug transporters by PCN in WT and PXR-null mice to determine which transporters are regulated by PCN, not only in mouse liver, but also kidney, duodenum, jejunum, and ileum, and to determine whether induction by PCN is mediated by the PXR receptor.

Materials and Methods

Chemicals. Sodium chloride, HEPES sodium salt, HEPES free acid, lithium lauryl sulfate, EDTA, Pregnenolone-16 α -carbonitrile (PCN), and D-(+)-glucose were purchased from Sigma-Aldrich (St. Louis, MO). Micro-O-protect was purchased from Roche Bioscience (Indianapolis, IN). Formaldehyde, 4-morpholinepropanesulfonic acid (MOPS), sodium citrate, and NaHCO₃ were purchased from Fischer Scientific (Fairlawn, NJ). Chloroform, agarose, and ethidium bromide were purchased from AMRESCO Inc. (Solon, OH).

Chemical Treatments in Mice. Adult male C57BL/6 mice were purchased from Charles River Laboratories Inc. (Wilmington, MA). Breeding pairs of PXR-null mice in the C57BL/6 background were kindly provided by Dr. Frank J. Gonzalez (Guo et al., 2003), which were originally engineered by inserting neomycin phosphotransferase gene into exon 1 region of the PXR gene (Staudinger et al., 2001b). Mice were maintained in a 12-hr dark/light cycle, temperature-, and humidity-controlled environment according to American Animal Associations Laboratory Animal Care guidelines and allowed water and rodent chow (Teklad; Harlan, Indianapolis, IN) ad libitum. Groups of five mice (WT, WT+PCN, PXR-null, and PXR-null+PCN) were administered once daily (10 ml/kg of body weight) for 4 days either corn oil or PCN (200 mg/kg, ip in corn oil). Liver, kidney, and 3 sections of small intestine (duodenum, jejunum, and ileum) were removed on day 5, snap-frozen in liquid nitrogen, and stored at -80°C.

Total RNA Isolation. Total RNAs were isolated using RNA Bee reagent (TelTest Inc., Friendswood, TX) per the manufacturer's protocol. The concentration of total

RNA in each sample was quantified spectrophotometrically at 260 nm. The integrity of each RNA sample was evaluated by formaldehyde-agarose gel electrophoresis before analysis.

Development of Specific Oligonucleotide Probe Sets for Branched DNA

(bDNA) Analysis. Gene sequences of interest were accessed from GenBank.

bDNA probe set design for each gene has been described previously (Cheng et al., 2005a). Probes were synthesized by QIAGEN Operon (Alameda, CA). Probe sets for mouse Cyp3a11 (Cheng et al., 2005c), Oatp1a4 (Cheng et al., 2005a), Mrp2 and 3 (Maher et al., 2005b), Cnt1 and 2 (Lu et al., 2004), and Abca1 (Dieter et al., 2004) have been reported. Probe sets for mouse PXR, Mdr1a, Bsep, and Ibat, as well as other transporter genes are shown in the Supplement (Table s1).

bDNA Assay. Reagents required for RNA analysis (i.e., lysis buffer, amplifier/label probe buffer, and substrate solution) were supplied in the Quantigene[®] bDNA signal amplification kit (Bayer Diagnostics, East Walpole, MA). The mRNA level of each gene were analyzed as described previously (Hartley and Klaassen, 2000). Pooled total RNA samples from 5 mice were used to determine the effects of PCN on transporter expression. Apparent altered levels of transporter mRNA were confirmed by quantifying individual mouse total RNA samples by the bDNA technique. Data are presented as relative light units (RLUs) per 10 µg total RNA.

Statistical Analysis. When individual RNA samples were used, data were expressed as mean ± S.E.M. Data were analyzed by one-way ANOVA, followed by Duncan's post-hoc test. Statistical significance was set at $P < 0.05$.

Results

Regulation of mouse PXR mRNA expression by PCN. The major organs in the body important for drug metabolism and excretion include liver, kidney, and small intestine. In this study, PXR mRNA was shown to be highly expressed in small intestine, modest in liver, and least in kidney (Fig. 1a). PCN is a well-known rodent PXR ligand and activator. In the present study, PCN did not alter the expression of PXR mRNA in mouse liver, kidney, or small intestine (Fig. 1a).

Regulation of mouse Cyp3a11 mRNA expression by PCN. Cyp3a is a well-characterized PXR-target gene (Lehmann et al., 1998; Masuyama et al., 2001; Cheng et al., 2005c). As shown in Fig. 1b, Cyp3a11 mRNA has the highest expression in mouse liver, modest in duodenum and jejunum, and least in kidney and ileum. PCN induced Cyp3a11 mRNA expression 2.2-, 2.3-, 2.9-, and 4.8-fold in liver, duodenum, jejunum, and ileum, respectively; whereas in kidney, PCN did not alter Cyp3a11 mRNA expression. In PXR-null mice, PCN did not increase Cyp3a11 mRNA level in any of the tissues.

Regulation of major hepatic transporters by PXR and PCN. In liver, multiple transporters are responsible for uptake and efflux of xenobiotics from hepatocytes. Regulation of hepatic transporters by PCN in WT and PXR-null mice is illustrated in Fig. 2. Among 19 transporters tested, elimination of the PXR gene in mouse liver decreased constitutive mRNA levels of Oatp1a4 (83%) and Bsep (40%). PCN induced the uptake transporter Oatp1a4 mRNA expression about 930% and the efflux transporter Mrp3 mRNA expression about 230% in

WT mice, but not in PXR-null mice. PCN treatment did not alter the mRNA expression of any other transporter in mouse liver.

Regulation of intestinal transporters by PXR and PCN. The small intestine is important for the absorption of orally administered pharmaceuticals and other compounds. In addition to passive diffusion, transporter systems are involved in absorption. Some transporters, such as Pept1, Cnt2, Bcrp, and Mrp2 are highly expressed in small intestine. A total of 18 transporters were investigated (see Supplement Figs s3-s5). However, the expression of only a few transporters was altered in mouse duodenum, jejunum, and ileum by PCN treatment and/or PXR disruption, as shown in Fig. 3.

In duodenum, removal of functional PXR protein, as illustrated in the PXR-null mice, increased constitutive mRNA level of the uptake transporter Cnt1 (75%), and the efflux transporters Mdr1a (150%) and Mrp2 (100%). PCN treatment increased duodenal Abca1 (50%) and Mdr1a (150%) mRNA level, but not in PXR-null mice.

In jejunum, disruption of PXR increased constitutive mRNA levels of Cnt1 (130%) and Mrp2 (110%), but decreased constitutive mRNA levels of Cnt2 about 55%. PCN induced mRNA expression of efflux transporters Abca1 (50%), Mdr1a (90%), and Mrp2 (50%), whereas it decreased the mRNA of the uptake transporter Cnt2 (40%) in jejunum. However, in jejunum of PXR-null mice, PCN did not change Abca1, Mdr1a, Mrp2, and Cnt2 mRNA expression, but PCN increased Cnt1 mRNA in PXR-null mice.

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In ileum, disruption of PXR increased constitutive mRNA expression of Mrp2 (80%), but decreased constitutive mRNA levels of Cnt2 (47%). PCN increased mRNA transcripts of efflux transporters Mdr1a (70%) and Mrp2 (160%), whereas PCN decreased the mRNA expression of the uptake transporter Cnt2 about 30%. However, in ileum of PXR-null mice, PCN did not alter Mdr1a, Mrp2, and Cnt2 mRNA expression, but decreased Ibat mRNA abundance about 55%.

Discussion

The major organs in the body important for drug disposition include liver, kidney, and small intestine. It has been shown that mouse PXR mRNA is expressed in liver, intestine, uterus, ovary, and placenta (Masuyama et al., 2001). In this study, PXR mRNA was shown to be highly expressed in small intestine, modest in liver, and least in kidney (Fig. 1a). Therefore PXR is more likely involved in regulating drug metabolism and excretion in small intestine and liver, than in kidney. In the present study, PXR and PCN regulate the expression of Cyp3a11 and some transporters in mouse liver and intestine. However, neither elimination of PXR nor PCN administration appeared to alter mRNA expression of any transporter in mouse kidney (Data are shown in the Supplement: Fig. s2).

PCN is a well-known rodent PXR activator. In mouse lung, PCN has previously been shown not to alter PXR mRNA expression (Haag et al., 2003). In the present study, PCN did not alter the expression of PXR in mouse liver, kidney, or small intestine (Fig. 1a). Therefore, PCN does not alter PXR mRNA expression in mice.

Cyp3a is a well-characterized PXR-target gene (Lehmann et al., 1998; Masuyama et al., 2001; Cheng et al., 2005c). As reported previously, PCN induces Cyp3a11 in mouse liver and intestine by a PXR-dependent mechanism (Fig. 1b) (Hsiang et al., 1999; Masuyama et al., 2001; Staudinger et al., 2001b; Matheny et al., 2004).

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Regulation of Cyp3a and some transporters by PXR agonists varies between species. As stated previously, PCN induced Cyp3a11 in both mouse liver and intestine by a PXR-dependent mechanism. However in rats, PXR agonists L-742694 and DEX increased Cyp3a1/23 and Cyp3a18 in liver, but decreased Cyp3a1/23 and Cyp3a18 in intestine (Hartley et al., 2004). In humans, PXR agonist rifampicin induced CYP3A4 and CYP3A5 expression in both human liver and intestine (Greiner et al., 1999; Burk et al., 2004). Therefore, Cyp3a/CYP3A regulation is similar in liver between rodents and humans, whereas their regulation in intestine is variable between species. Another example is the regulation of Mrp2 in mouse and rat intestine by PXR agonists. In female rat intestine, L-742694 did not alter Mrp2 mRNA expression, and DEX decreased intestinal Mrp2 mRNA level to half that of control (Hartley et al., 2004). However, in mouse intestine, PCN increased Mrp2 mRNA abundance, as shown in Fig 3. The chemical L-742694 might have properties in addition to being a PXR ligand, and these other functions may be involved in the regulation of these target genes (Hartley et al., 2004). However, PCN is a quite specific rodent PXR activator. Therefore, regulation of gene expression by L-742694 may be due to activation of multiple transcription factors, whereas regulation by PCN is solely via PXR. In addition, Abcg5 and Abcg8, two important sterol efflux transporters, are also regulated differently in rats and mice by PCN. In rats, PCN treatment induces hepatic Abcg5 and Abcg8 mRNA expression (Dieter et al., 2004). However, in mouse liver and intestine, PCN

treatment did not alter Abcg5 and Abcg8 mRNA expression (data shown in the Supplement: Figs. s1, s3-s5).

As shown in Figs 2 and 3, PCN increases Oatp1a4 and Mrp3 mRNA in liver, as well as Mdr1a and Mrp2 mRNA in small intestine of WT mice, but not in PXR-null mice, indicating that PCN induction of these transporters is PXR-dependent. In addition to up-regulation of some transporters, PCN administration also decreased Cnt2 mRNA in jejunum and ileum of WT mice, but not in PXR-null mice, indicating that the decrease is mediated via PXR. Regulation of rat and mouse hepatic Oatp1a4 (Staudinger et al., 2001a; Guo et al., 2002c) and Mrp3 (Teng et al., 2003), as well as mouse intestinal Mdr1a (Geick et al., 2001; Matheny et al., 2004) by PCN has been reported previously. The present study confirms that mouse hepatic Oatp1a4 and Mrp3, and intestinal Mdr1a regulation by PCN is PXR-dependent.

PXR is required for the induction of some Cyps and transporters by PCN, but also is involved in the constitutive expression of some genes. After disruption of PXR, as in PXR-null mice, constitutive expression of Oatp1a4 and Bsep in liver; Cnt1, Mdr1a, and Mrp2 in duodenum; Cnt1, Cnt2, and Mrp2 in jejunum; as well as Cnt2 and Mrp2 in ileum are altered. Whereas Cnt2 mRNA is decreased in PXR-null mice, mRNA of other transporter genes is increased in the PXR-null mice. Therefore, PXR regulates genes at two different levels, constitutive expression and ligand-induced expression.

PCN has been historically classified as a "catatoxic" or protective steroid, which protects against the toxic effects of various chemicals, presumably by

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inducing liver biotransformation enzymes (Solymoss et al., 1969). PCN acts via PXR to increase Cyp3a11 mRNA level in all five tissues, the uptake transporter Oatp1a4 and efflux transporters Mrp3 in liver, as well as Mdr1a and Mrp2 in small intestine. As a result, activation of PXR will (1) induce intestinal efflux transporters Mdr1a and Mrp2, and thus decrease bioavailability of some xenobiotics by transporting them back into the intestinal lumen; (2) induce Cyp3a11 in liver, kidney, and intestine and thus enhance the biotransformation of some xenobiotics; (3) induce liver uptake transporter Oatp1a4 and efflux transporters Mrp3, leading to enhanced liver extraction and biotransformation, with subsequent transport into the blood for excretion by the kidney. Therefore, the catatoxic steroid PCN, via PXR, can protect the body against xenobiotic toxicity by both enhancing biotransformation via Cyp3a, and accelerating the transport of harmful chemicals from the body, as reported previously (Klaassen, 1974a; Kourounakis et al., 1977; Gregus et al., 1990; Chen et al., 2003).

PCN may enhance intestinal absorption of dietary cholesterol and other nutrients. Whole-body cholesterol homeostasis is stringently regulated by de novo synthesis, dietary cholesterol absorption, and biliary excretion. In the intestine, Niemann-pick C1 like 1 transporter (Npc1L1) is critical for the apical transport of cholesterol from the lumen across the apical membrane into enterocytes (Altmann et al., 2004), whereas Abca1 is responsible for basolateral transport of cholesterol from the enterocyte into the bloodstream (Mulligan et al., 2003). In the present study, Npc1L1 is highly expressed in intestine and PCN does not alter its expression (data shown in the Supplement: Figs s3-s5),

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whereas Abca1 is moderately expressed in intestine and PCN increases Abca1 mRNA level (Fig 3). Therefore, PCN may increase intestinal cholesterol absorption by enhancing cholesterol efflux from enterocytes into blood.

PXR-regulated transporter expression could result in drug-drug interactions. The ligands that bind to PXR are environmental chemicals, pharmaceuticals, and endogenous compounds. For example, rodent PXR is activated by PCN, dexamethasone (DEX), spironolactone, bile acids (Staudinger et al., 2001b; Frank et al., 2005), and other compounds (Guo et al., 2002a; Cheng et al., 2005c). Human PXR is activated by pharmaceuticals such as rifampicin, clotrimazole, phenobarbital, nifedipine, RU486 (mifepristone), metyrapone, and hyperforin, as well as other compounds such as the bile acids lithocholic acid and 6-keto lithocholic acid (Waxman, 1999; Moore et al., 2000). Furthermore, a number of pharmaceuticals are substrates for the transporters that are regulated by PXR. For example, Oatp1a4 can transport the cardiac glycosides digoxin and ouabain, the antihistamine fexofenadine, and the cholesterol-lowering drug pravastatin (Hagenbuch and Meier, 2003); Mdr1 transports paclitaxel, vinblastine, ivermectin, digoxin, cyclosporin A, rifampicin, and dexamethasone (Brady et al., 2002); whereas Mrp2 and 3 transport anticancer drugs anthracyclines, vinca alkaloids, and methotrexate (Maher et al., 2005b). These broad substrate specificities and the ability to alter transporter expression through PXR increase the likelihood that when two or more pharmaceuticals are used at the same time, the potential for drug-drug interactions at the level of transporters may affect pharmacokinetics. Cnt2 mRNA abundance is decreased

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by PCN in mouse jejunum and ileum, which is important in the intestinal absorption and disposition of naturally occurring nucleosides, as well as nucleoside-derived drugs used in antiviral and cancer chemotherapies. In addition, PCN also induced mRNA expression of Mdr1a, an efflux transporter, which is capable of transporting substrates out of enterocytes into the intestinal lumen. Therefore, PXR activators may reduce efficacy of some oral anticancer therapeutics by decreasing intestinal uptake via Cnt2 and enhancing intestinal excretion by Mdr1a.

The present study indicates that the regulation of PXR-targeting genes in various tissues is not identical. For example, Mrp2 is not altered by PCN in mouse liver at the mRNA level, but induced in small intestine by PCN in a PXR-dependent manner. The present data are in accordance with a recent report (Maher et al., 2005a) that showed Mrp2 is not induced by PCN in mouse liver. However, it has been reported that Mrp2 may be regulated in rat liver at a post-translational step (Gerk and Vore, 2002; Johnson and Klaassen, 2002; Jones et al., 2005). Mdr1a is induced in mouse small intestine by PCN, but not in either liver or kidney. This indicates that PCN via PXR can regulate some target genes in one tissue, but not in another tissue, where there is a high level of PXR.

The regulation of transporters can theoretically occur at the transcriptional and/or translational level. Due to the limited availability of good antibodies and specific substrates for each transporter, regulation of transporters has mainly been determined at the mRNA level. However, the increase in expression of

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Oatp1a4 mRNA correlates with a marked increase in clearance of ouabain by the liver (Klaassen, 1974b; Eaton and Klaassen, 1978; Guo et al., 2002c).

In conclusion, the PXR ligand PCN functions as a "catatoxic" or protective steroid, by enhancing presystemic elimination of compounds, including both enhancing Cyp3a biotransformation enzymes and accelerating the transport of harmful chemicals from the body. However, the benefits of efficient detoxification may be complicated by potential drug-drug interactions.

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Footnotes

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Figure legends

Fig. 1 a. Expression of PXR mRNA in liver, kidney, duodenum, jejunum, and ileum of male C57BL/6 mice. b. Expression of Cyp3a11 mRNA in liver, kidney, duodenum, jejunum, and ileum of male C57BL/6 and PXR-null mice after administration of pregnenolone-16 α -carbonitrile (PCN). Black bars represent WT mouse tissues; grey bars represent PCN-treated WT mouse tissues; latticed bars represent PXR-null mouse tissues; striated bars represent PCN-treated PXR-null mouse tissues. Four treatment groups were conducted: C57BL/6 mouse, C57BL/6 mouse-PCN, PXR-null mouse, and PXR-null mouse-PCN. Mice (n=5) were administered once daily for 4 days either corn oil ip as the control group, or with PCN (200 mg/kg, ip in corn oil). After isolation of total RNA, individual RNA samples of 5 mice were analyzed by bDNA assay. Data were expressed as mean \pm S.E.M. for five mice in each group. Asterisk (*) indicates statistically significant difference between treated and control mice ($p < 0.05$). Dagger (†) indicates a statistically significant difference in constitutive transporter mRNA expression between C57BL/6 and PXR-null mice.

Fig. 2 mRNA expression of Oatp1a4, Bsep, and Mrp3 transporters in livers of male C57BL/6 and PXR-null mice after administration of pregnenolone-16 α -carbonitrile (PCN). Black bars represent WT mouse tissues; grey bars represent PCN-treated WT mouse tissues; latticed bars represent PXR-null mouse tissues; striated bars represent PCN-treated PXR-null mouse tissues. Four treatment groups were conducted: C57BL/6 mouse, C57BL/6 mouse-PCN,

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PXR-null mouse, and PXR-null mouse-PCN. Mice (n=5) were administered once daily for 4 days either corn oil ip as the control group, or with PCN (200 mg/kg, ip in corn oil). After isolation of total RNA, pooled RNA samples or individual RNA samples of 5 mouse livers were analyzed by bDNA assay. Where individual RNA samples were quantified, data were expressed as mean \pm S.E.M. for five mice in each group. Asterisk (*) indicates statistically significant difference between treated and control mice ($p < 0.05$). Dagger (†) indicates a statistically significant difference in constitutive transporter mRNA expression between C57BL/6 and PXR-null mouse livers.

Fig. 3 mRNA expression of Cnt1/2, Ibat, Mdr1a, Mrp2, and Abca1 transporters in duodenum, jejunum, and ileum of male C57BL/6 and PXR-null mice after administration of pregnenolone-16 α -carbonitrile (PCN). Black bars represent WT mouse tissues; grey bars represent PCN-treated WT mouse tissues; latticed bars represent PXR-null mouse tissues; striated bars represent PCN-treated PXR-null mouse tissues. Four treatment groups were conducted: C57BL/6 mouse, C57BL/6 mouse-PCN, PXR-null mouse, and PXR-null mouse-PCN. Mice (n=5) were administered once daily for 4 days either corn oil ip as the control group, or with PCN (200 mg/kg, ip in corn oil). Data are expressed as mean \pm S.E.M. for five mice in each group. Asterisk (*) indicates statistically significant difference between treated and control mice ($p < 0.05$). Dagger (†) indicates statistically significant difference in constitutive transporter mRNA expression between C57BL/6 and PXR-null mouse duodenum.

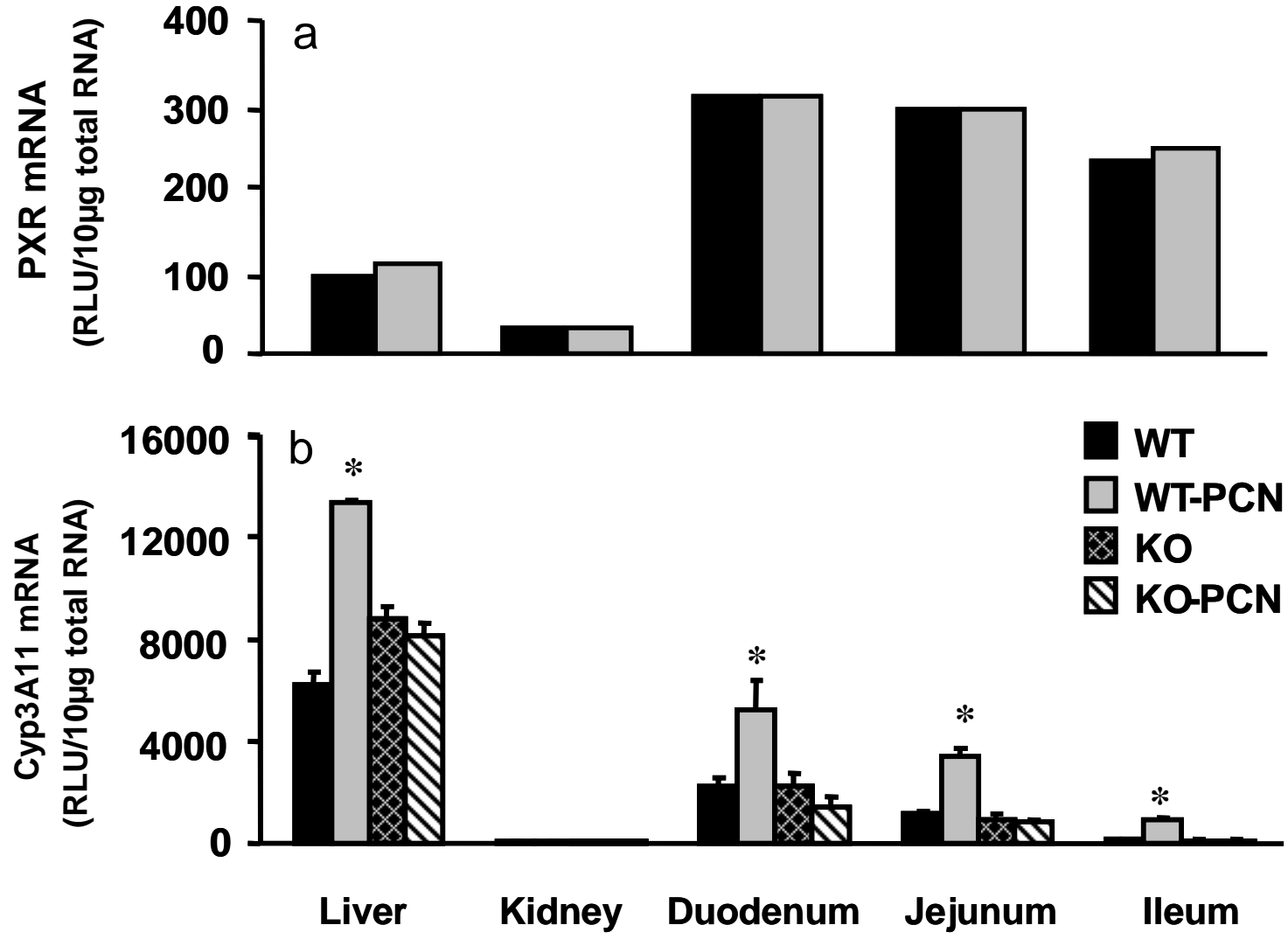


Fig. 1

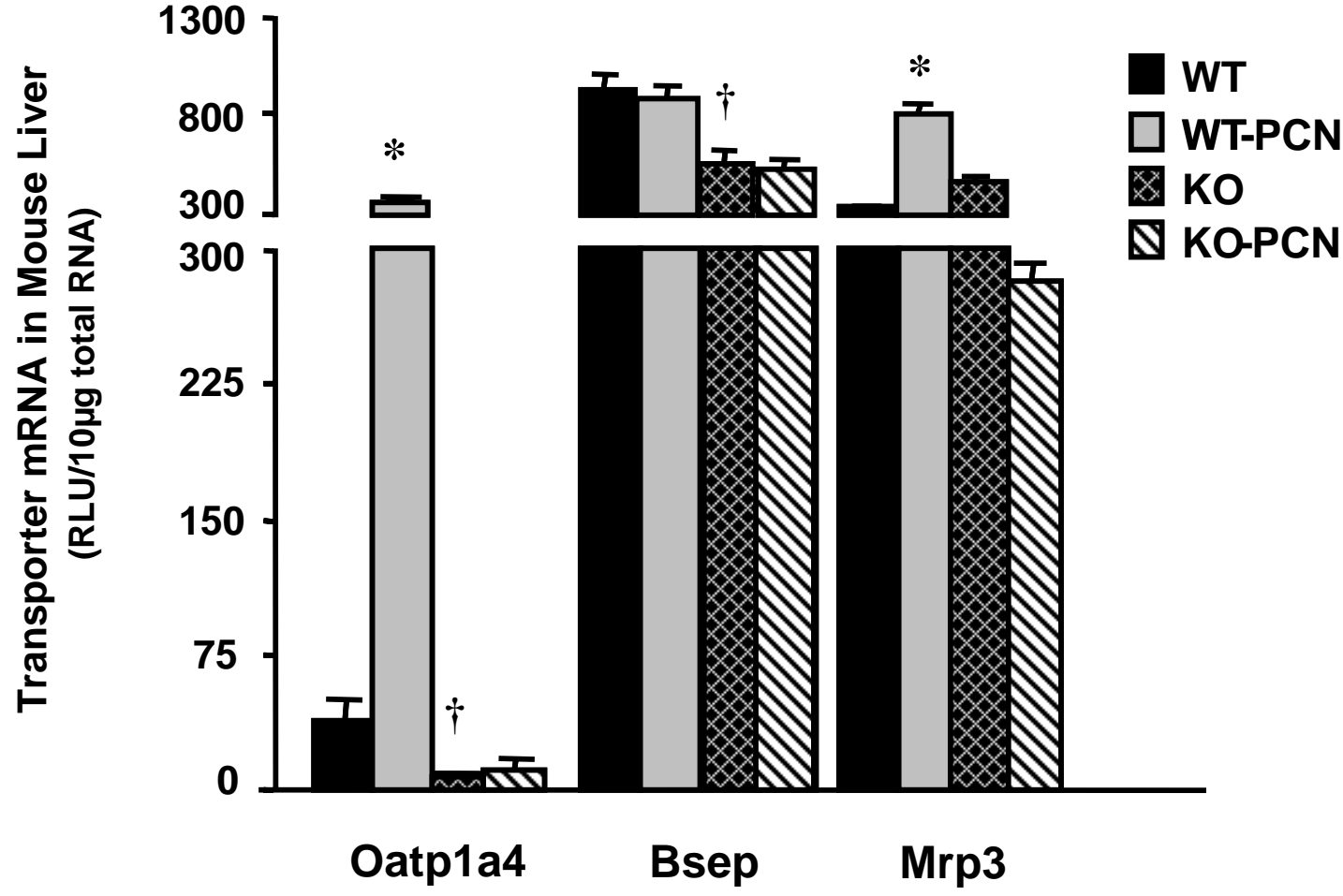


Fig. 2

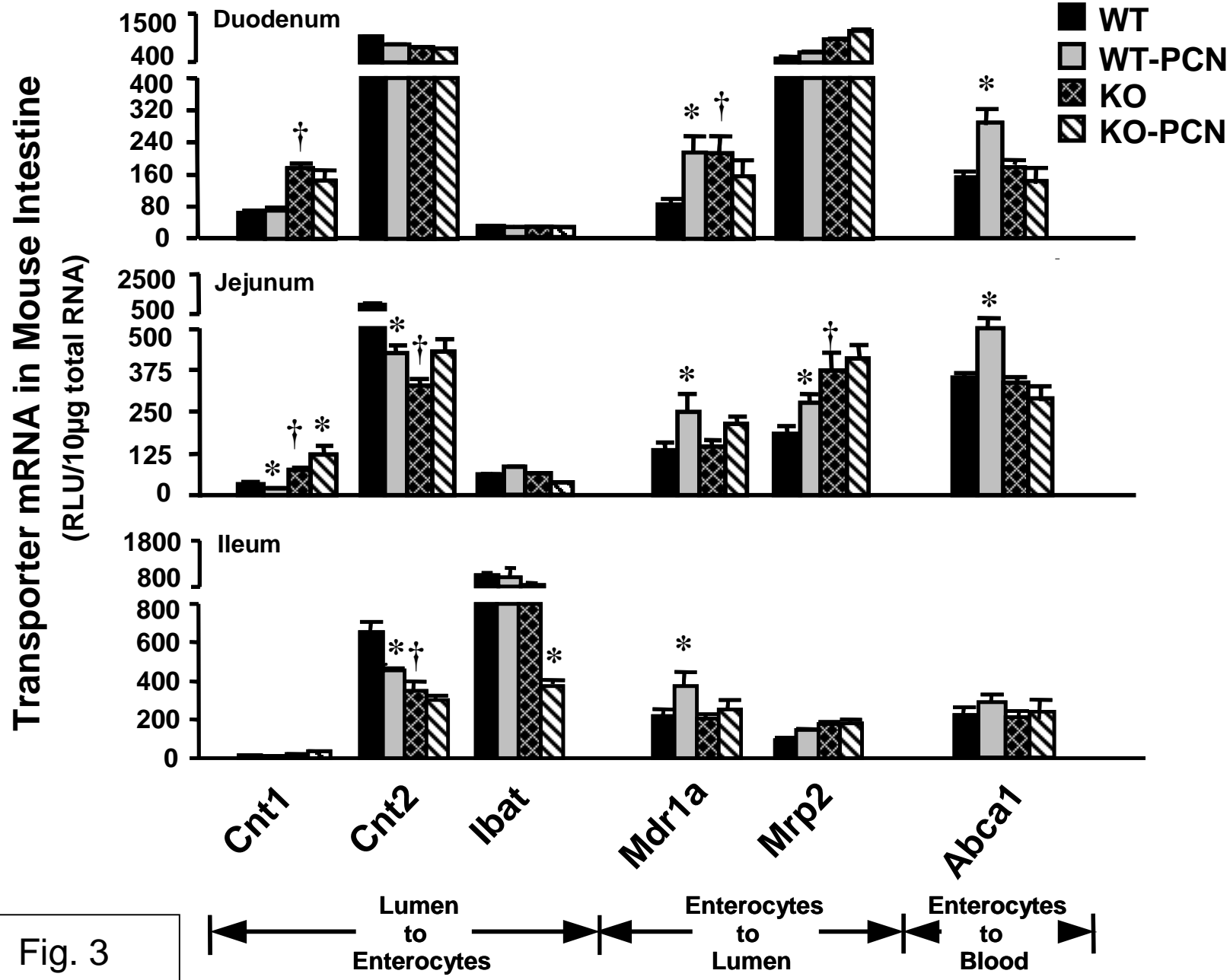


Fig. 3