Tissue- and Gender-Specific mRNA Expression of UDP-Glucuronosyltransferases (UGTs) in Mice

David B. Buckley and Curtis D. Klaassen

Department of Pharmacology, Toxicology, and Therapeutics, University of Kansas Medical Center, Kansas City, Kansas

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ABSTRACT:

UDP-glucuronosyltransferases (UGTs) catalyze phase II biotransformation reactions in which lipophilic substrates are conjugated with glucuronic acid to increase water solubility and enhance excretion. Currently, little information regarding tissue- or gender-specific expression of mouse UGTs is available. Mice are increasingly popular models in biomedical research, and therefore, thorough characterization of murine drug metabolism is desired. The purpose of the present study was to determine both tissue- and gender-specific UGT gene expression profiles in mice. RNA from 14 tissues was isolated from male and female C57BL/6 mice and UGT expression was determined by the branched DNA signal amplification assay. UGTs highly expressed in mouse liver include Ugt1a1, Ugt1a5, Ugt1a6, Ugt1a9, Ugt2a3, Ugt2b1, Ugt2b5/37/38, Ugt2b34, Ugt2b35, and Ugt2b36. Several isoforms were expressed in the gastrointestinal (GI) tract, including Ugt1a6, Ugt1a7c, Ugt2a3, Ugt2b34, and Ugt2b35. In kidney, Ugt1a2, Ugt1a7c, Ugt2b5/37/38, Ugt2b35, and Ugt3a1/2 were expressed. UGT expression was also observed in other tissues: lung (Ugt1a6), brain (Ugt2b35), testis and ovary (Ugt1a6 and Ugt2b35), and nasal epithelium (Ugt2a1/2). Male-predominant expression was observed for Ugt2b1 in liver, Ugt2b5/37/38 in kidney, and Ugt1a6 in lung. Female-predominant expression was observed for Ugt1a1 and Ugt1a5 in liver, Ugt1a2 in kidney, Ugt2b35 in brain, and Ugt2a1/2 in nasal epithelium. UDP-glucose pyrophosphorylase was highly expressed in liver, kidney, and GI tract, whereas UDP-glucose dehydrogenase was highly expressed in the GI tract. In conclusion, marked differences in tissue- and gender-specific expression patterns of UGTs exist in mice, potentially influencing drug metabolism and pharmacokinetics.

UDP-glucuronosyltransferases (UGTs) catalyze phase II biotransformation reactions, conjugating lipophilic substrates with glucuronic acid, thereby increasing hydrophilicity and enhancing excretion through bile and urine. Glucuronidation reactions are generally considered detoxification reactions, in which a potentially toxic substrate is conjugated to glucuronic acid. The addition of the glucuronic acid moiety occurs at hydroxyl, carboxylic acid, amine (primary, secondary, and tertiary), carbonyl, and thiol groups (Dutton, 1980). Endogenous UGT substrates comprise numerous steroids and metabolic by-products, including bilirubin, testosterone, thyroxine, estradiol, androsterone, and hyodeoxycholic acid. In addition, numerous xenobiotics serve as UGT substrates, including drugs such as acetaminophen, morphine, propofol, chloramphenicol, and nonsteroidal anti-inflammatory drugs, as well as environmental compounds, such as plant-derived dietary flavonoids, carcinogens, and other pollutants (Dutton, 1980; Radominska-Pandya et al., 1999; Tukey and Straussburg, 2000).

The UGT superfamily of genes is divided into two families, UGT1 and UGT2, based on sequence similarity at the amino acid level. UGT1 family genes are encoded from a unique first exon (I) spliced with common exons 2 to 5 (II–V), creating a common C-terminal domain and unique N-terminal domains; each UGT1A family member contains gene-specific promoter regions (Mackenzie et al., 1997, 2005). In humans, 13 unique first exons have been identified (UGT1A1–13), encoding nine functional enzymes and four pseudogenes (UGT1A2, 11, 12, 13), whereas in rats, nine first exons have been identified (UGT1A1–9), with two coding pseudogenes (UGT1A4 and UGT1A9) (Emi et al., 1995; Gong et al., 2001). In mice, the Ugt1a subfamily contains 14 first exons, coding nine enzymes (Ugt1a1, 2, 5, 6, 6a, 7a, 7c, 8, 9, and 10) and five pseudogenes (Ugt1a3, 4, 7a, 7b, and 11) (Mackenzie et al., 2005).

Unlike UGT1 genes, each UGT2 gene is encoded by six individual exons and is further divided into two families, UGT2A and UGT2B. UGT2A1 mRNA is expressed in the nasal epithelia of both humans and rats (Jedlitschky et al., 1999; Heydel et al., 2001). Recent reports indicate that the gene duplication product of UGT2A1 in humans, UGT2A2, is expressed in liver (Jedlitschky et al., 1999; Heydel et al., 2001; Tukey and Straussburg, 2001). Analysis of the mouse genome indicates that there are three Ugt2a gene duplication products, Ugt2a1, Ugt2a2, and Ugt2a3 (Mackenzie et al., 2005). Humans, rats, and mice each contain seven unique UGT2B genes (Mackenzie et al., 2005). The seven Ugt2b genes in mice include Ugt2b1, 2b5, 2b34, 2b35, 2b36, 2b37, and 2b38. In addition, a novel UGT3A family has been identified, which contains gene-specific promoter regions (Mackenzie et al., 1997, 2005). The seven Ugt3a genes in mice include Ugt3a1, 2, 3, 8, 9, and 10 (Mackenzie et al., 2005).

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ABBREVIATIONS: UGT, uridine diphosphate glucuronosyltransferase; UDP-GA, uridine diphosphate glucuronic acid; UDP-gpp, UDP-glucose pyrophosphorylase; UDP-gdh, UDP-glucose dehydrogenase.
been identified in humans and rodents, giving rise to two individual gene products in mice, Ugt3a1 and Ugt3a2, located on mouse chromosome 15 (Mackenzie et al., 2005). In general, UGT1A genes share homology between species (human UGT1A1 is homologous to rat and mouse Ugt1a1). Conversely, the UGT2B family does not share orthologs between species, except for UGT2B1 in rats and mice (Mackenzie et al., 2005).

Expression of UGT mRNA and protein, and thus, glucuronidation, occurs primarily in liver, gastrointestinal tract, and kidney. Human UGT1A transcripts are highly expressed in liver, kidney, and portions of the GI tract, including gastric tissue, small intestine, and colon, which are primarily responsible for glucuronidation of dietary substances and drugs (Mojarra and Mackenzie, 1998; Strassburg et al., 1998a,b, 2000; Fisher et al., 2001; Basu et al., 2004). Likewise, mRNA expression of rat UGT1A isoforms is predominant in liver and intestine (Emi et al., 1995; Grams et al., 2000; Shelby et al., 2003). In mice, several Ugt1a transcripts have been detected in liver, kidney, stomach, esophagus, small intestine, and colon (Zhang et al., 2004).

However, human and rat UGT2B members are expressed not only in liver and intestine, but also in steroid-target tissues such as testis, uterus, mammary tissue, and brain, where they conjugate numerous endogenous steroids (Beaulieu et al., 1996, 1998; Levesque et al., 1997, 1999; Turgeon et al., 2001; Shelby et al., 2003). Hence, tissue-specific expression of individual UGT members contributes to the organism’s ability to selectively and preferentially glucuronidate chemicals in specific tissues.

Gender differences in UGT expression and function have been documented in rats. In Wistar rats, the rate of bilirubin glucuronidation in liver microsomes is 2-fold higher in females than in males (Muraca and Fevity, 1984). Castration of male rats increased bilirubin glucuronidation, and androgen treatment reduced the rate of bilirubin glucuronide formation. Ovariectomized female rats displayed a decreased bilirubin glucuronidation rate, whereas treatment with progestins increased glucuronidation rates (Muraca and Fevity, 1984). Expression of rat UGT2B3 mRNA was higher in males than in females, and could be reduced by castration and restored by treatment with testosterone (Strasser et al., 1997). These examples illustrate the importance of understanding gender-specific UGT expression to determine gender-specific glucuronidation profiles.

The tissue distribution of mouse Ugt1a members has been examined (Zhang et al., 2004). However, gender differences in mouse Ugt1a gene expression have not been explored. Also, mouse Ugt2 and Ugt3 tissue- and gender-specific expression patterns have not been examined. Therefore, the present study determines the tissue- and gender-specific mRNA expression patterns of mouse UGTs as well as the enzymes responsible for the synthesis of UDP-glucuronic acid (UDP-GA).

**Materials and Methods**

**Animals.** Male and female C57BL/6 mice (n = 5 per gender) 8 weeks of age were purchased from Charles River Laboratories, Inc. (Wilmington, MA). Animals were housed according to the American Animal Association Laboratory Animal Care guidelines. Mice were allowed food (Harlan-Teklab) and water ad libitum, and were acclimated to the housing facility for 1 week. Twelve tissues were removed, including liver, kidney, stomach, duodenum, jejunum, ileum, large intestine, lung, brain, heart, and gonads (ovaries and testes). Ovaries were pooled (25 ovaries for n = 1), and additional ovaries were collected from timed-pregnant females at gestation day 18. Nasal efflatory epithelia were collected and pooled into a single sample for each gender (n = 1/gender). Stomach and intestine were dissected and rinsed in saline. All tissues were snap-frozen in liquid nitrogen and stored at -80°C.

**Total RNA Isolation.** Total RNA was extracted from each tissue using RNA-Bee Reagent (Tel-Test Inc., Friendswood, TX) according to the manufacturer’s protocol. RNA was quantified by UV spectrophotometry at 260/280 nm and diluted to 1 μg/μl in diethyl pyrocarbonate-treated water. RNA samples were analyzed by formamide-agarose gel electrophoresis, and integrity was confirmed by visualization of 18S and 28S rRNA bands.

**Development of Oligonucleotide Probe Sets for bDNA Analysis.** Multiple oligonucleotide probe sets (capture extender, label extender, and blocker probes) were designed to mouse UGT cDNA sequences obtained from GenBank. Probe sets for Ugt1a1, 1a2, 1a6, and 2b5 were designed previously (Chen et al., 2003). Due to high DNA sequence similarity of Ugt2b5, 2b37, and 2b38, the Ugt2b5 oligonucleotide probe set designed for Ugt2b5 by Chen et al. (2003) has a high probability of cross-reacting, and has been given the annotation Ugt2b5/37/38 to note this. Probe sets for mouse Ugt1a5, 1a7c, 1a8, 1a9, 1a10, 2a1/2, 2a3, 2a6, 2b34, 2b35, 2b36, and 3a1/2 were designed by the following methods. To determine the minimum cross-reactivity with other mouse sequences, each sequence was submitted to BLASTn for nucleotide comparison. Probes with >80% sequence homology to other mouse sequences were eliminated from the probe design. Probe sets were synthesized by QIAGEN Operon (Alameda, CA) or Integrated DNA Technologies, Inc. (Coralville, IA) and diluted in 1× Tris-EDTA buffer (pH 8.0) and stored at -20°C. Similar to the Ugt2b5/37/38 probe set, sequence homology required probe sets for Ugt2a1/2 and Ugt3a1/2 to cross-react with, and detect, two genes; probe sets are named accordingly. A list of the newly designed oligonucleotide probe sets is located in Supplemental Data.

**bDNA Signal Amplification Assay.** Individual mouse UGT mRNA transcripts were detected using the Quantigene bDNA signal amplification assay (Panomics [formerly Genospetra Inc.], Fremont, CA). The bDNA assay was performed as described and validated previously (Hartley and Klaassen, 2000). In brief, capture extender, label extender, and blocker probes were combined and diluted into lysis buffer. Total RNA (1 μg/μl; 10 μl) was added to each well of 96-well plates containing 50 μl of capture hybridization buffer and 50 μl of diluted probe set, and allowed to hybridize at 53°C overnight. Plates were cooled to 46°C and rinsed twice with wash buffer. Amplifier reagent (10 μl), diluted 1:1000 in amplifier/label probe buffer, was added to each well and incubated at 46°C for 1 h. Plates were rinsed again with wash buffer and label

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**TABLE 1**

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*Based 91.75-92.580.
Results

Tissue Distribution of Mouse Ugt1a Subfamily Genes. The tissue- and gender-specific mRNA expression of Ugt1a1, 1a2, 1a5, and 1a6 genes is illustrated in Fig. 1. Ugt1a1, the bilirubin-conjugating isozyme, mRNA was highly expressed in liver in a female-predominant pattern (Fig. 1). Ugt1a1 mRNA expression was also observed at low levels in duodenum, jejunum, ileum, and large intestine, whereas expression was negligible in all other tissues examined. Ugt1a2 mRNA was highly and almost exclusively expressed in female kidney (Fig. 1). Ugt1a5 mRNA transcripts were expressed in a pattern similar to that of Ugt1a1. Ugt1a5 mRNA was expressed highest in liver, where expression was predominant in females (Fig. 1). Low levels of Ugt1a5 were observed in lung and gastrointestinal tract, where males expressed more Ugt1a5 in jejunum than females. Expression of Ugt1a5 was observed at considerably lower levels in kidney, brain, gonads, and placenta. The phenol-conjugating isozyme, Ugt1a6, mRNA was more ubiquitously expressed (Fig. 1). Ugt1a6 mRNA was expressed to the highest extent in liver and large intestine, followed by lung, stomach, and gonads. Low levels of Ugt1a6 were also detected in kidney and small intestine. In addition, a gender difference was also observed for Ugt1a6 in lung, where males expressed higher levels than female mice.

The tissue- and gender-specific mRNA expression profiles of mouse Ugt1a7c, 1a8, 1a9, and 1a10 are illustrated in Fig. 2. Ugt1a7c mRNA was primarily expressed in intestine and kidney (Fig. 2). Ugt1a7c transcripts were highest in large intestine, followed by duodenum, kidney, jejunum, ileum, and stomach. No gender differences were observed for mouse Ugt1a7c. Ugt1a8 was expressed at very low levels in all mouse tissues examined, exhibiting high background readings, raising concerns whether Ugt1a8 mRNA is truly expressed in tissues examined (Fig. 2). Ugt1a9 mRNA was mainly expressed in liver, with very low levels detected in the other tissues examined (Fig. 2). Hepatic Ugt1a9 expression tended to be higher in female than male mice; however, the difference was not statistically significant. Ugt1a10 was detected at low levels in several tissues (Fig. 2); most notable were kidney, liver, and stomach, where expression in kidney was female-predominant.

Tissue Distribution of Mouse Ugt2a Subfamily. Figure 3 illustrates the mRNA expression of the mouse Ugt2a subfamily. Ugt2a1/2 mRNA was highly and exclusively expressed in nasal epithelia in a female-predominant manner (Fig. 3). Ugt2a3 mRNA, like human UGT2A2, was detected at high levels in liver, with lower levels expressed in duodenum and jejunum (Fig. 3).

Tissue Distribution of Mouse Ugt2b Subfamily Genes. The tissue- and gender-specific expression of Ugt2b mRNAs is shown in Fig. 4. Mouse Ugt2b1, named for its orthology to rat UGT2B1, was highly and almost exclusively expressed in liver in a male-predominant pattern (Fig. 4). Ugt2b5/37/38 mRNA was detected at high levels in both liver and kidney (Fig. 4), whereas all other tissues examined had minimal expression. Ugt2b5/37/38 mRNA expression in kidney was higher in male than in female mice. In contrast to Ugt2b5/37/38,
Ugt2b34 transcripts were expressed at high levels in liver and the entire gastrointestinal tract (Fig. 4). Low levels of Ugt2b34 mRNA were also detected in kidney. Ugt2b35 was expressed, to some extent, in all tissues examined (Fig. 4). Specifically, Ugt2b35 mRNA was highly expressed in liver, stomach, and duodenum; moderately expressed in kidney, stomach, jejunum, ileum, and large intestine; and showed low expression in lung, heart, brain, gonads, and placenta. A gender difference was observed for Ugt2b35 in brain, where expression was higher in females than in male mice. Ugt2b36 mRNA, like Ugt2b1, was highly expressed in liver, with low expression levels in several tissues (Fig. 4).

**Tissue Distribution of Mouse Ugt3a Subfamily.** Ugt3a1/2 mRNA was highly expressed in kidney, was expressed at low levels in liver, and was not detected in other tissues examined (Fig. 5).

**Tissue Distribution of Mouse UDP-Glucose Pyrophosphorylase and UDP-Glucose Dehydrogenase.** Figure 6 illustrates the tissue- and gender-specific mRNA expression patterns of the two key enzymes responsible for UDP-glucuronic acid synthesis. UDP-glucose pyrophosphorylase (UDP-gpp) mRNA was highly expressed in liver, kidney, stomach, large intestine, and heart. Moderate expression was observed in brain and ovary. Lower levels of mRNA expression were detected in lung, duodenum, jejunum, ileum, and placenta. Females expressed higher levels of UDP-gpp in gonads than did males. UDP-glucose dehydrogenase (UDP-gdh) mRNA was highly expressed in the mouse gastrointestinal tract, including stomach, duodenum, jejunum, ileum, and large intestine. Lower levels of UDP-gdh mRNA were observed in liver, kidney, lung, heart, brain, gonad, and placenta. The observed level of UDP-gdh in liver was higher in male than in female liver. Conversely, UDP-gdh mRNA had higher expression in females in both brain and gonads.

**Discussion**

UDP-glucuronosyltransferases conjugate and generally detoxify various lipophilic endo- and xenobiotics to more hydrophilic com-
pounds that are readily excreted via bile and urine (Dutton, 1980). UGTs are expressed in a wide variety of tissues and are responsible for xenobiotic biotransformation and metabolism of endogenous compounds (Radominska-Pandya et al., 1999; Tukey and Strassburg, 2000; Turgeon et al., 2001). Therefore, glucuronidation undoubtedly influences the pharmacokinetic profiles of drugs in tissues with high glucuronidation capacity. Previous studies indicate that UGT mRNA expression, protein expression, and enzyme function correlate. In vitro, peroxisome proliferator-activated receptor α ligands increase human UGT2B4 mRNA expression as well as increase glucuronidation of bile acids (Barbier et al., 2003). Inducers of human UGT1A1, 1A4, and 1A6 mRNA and glucuronidation of bile acids (Barbier et al., 2003). Likewise, inducible transgenic mice, human UGT1A1, 1A4, and 1A6 mRNA and glucuronidation of bile acids (Barbier et al., 2003). In UGT human-UGT2B4 mRNA and protein expression as well as increase in vitro, peroxisome proliferator-activated receptor expression, protein expression, and enzyme function correlate. In human hepatocytes, both UGT1A1 and 1A4 mRNA and protein levels correlate in liver (Chen et al., 2005). Likewise, inducible transgenic mice, human UGT1A1, 1A4, and 1A6 mRNA and glucuronidation of bile acids (Barbier et al., 2003). In UGT humans and mice express UGT1A1, 1A6, and 1A9 mRNA in liver. Ugt1a5 and 1a9 in liver. For rodent-to-human comparisons, both humans and mice express UGT1A1, 1A6, and 1A9 mRNA in liver. Ugt1a6 in liver (Shelby et al., 2003), whereas mice also highly express Ugt1a6 in liver. The current data correspond to those in previous reports showing Ugt1a6 expression throughout the intestine (Zhang et al., 2004). However, Ugt1a6 was expressed highly in the large intestine, whereas levels were similar to that in liver. The current data correspond to those in previous reports showing Ugt1a6 expression throughout the intestine (Zhang et al., 2004). However, Ugt1a6 is highly expressed in large intestine in the current study, whereas Zhang et al., (2004) indicated that Ugt1a1 mRNA was expressed in colon, not Ugt1a6. In addition, only two Ugt2b transcripts, 2b34 and 2b35, were noticeably expressed in stomach as well as small and large intestine. Mouse Ugt1a7c and rat UGT1A7 share considerable expression similarities; both are predominantly expressed in small and large intestine (Shelby et al., 2003; Zhang et al., 2004), whereas human UGT1A7 is expressed in esophagus and stomach but not in intestine (Strassburg et al., 1998b). Both humans and mice express, to some extent, UGT1A1, 1A6, 1A7, and 1A10 along the gastrointestinal tract (Strassburg et al., 1998b). As for

### TABLE 2

Summary and comparison of the tissue distribution of human, rat, and mouse UGTs in liver, stomach, small intestine, colon, and kidney.

<table>
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<th>Rat*</th>
<th>Mouse</th>
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<td>a3; b1, 5, 34, 35, 36, 37, 38</td>
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<td></td>
<td>3A</td>
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<td>1, 2</td>
<td></td>
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<tr>
<td>Stomach</td>
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<tr>
<td></td>
<td>2B</td>
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<td>8</td>
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<tr>
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<td>2B</td>
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the UGT2B subfamily, UGT2B7, 2B10, and 2B15 are expressed in human esophagus (Tukey and Strassburg, 2000), whereas murine Ugt2b34 and 2B35 are present in stomach and large intestine.

Mouse Ugt1a8 expression was expected in intestine. However, Ugt1a8 was not detected above background signals in all tissues, raising concerns that mouse Ugt1a8 mRNA may not be expressed. These data are in agreement with previous reports failing to show Ugt1a8 mRNA expression (Zhang et al., 2004). In the present study, mouse Ugt1a8 probe set was designed to a portion of the mouse BAC RP23-396I23 (accession number AC087801; Bases: 91,725–92,580), which is highly homologous to rat UGT1A8. No expressed cDNAs or expressed sequence tags were found by BLASTn on the National Center for Biotechnology Information website (www.ncbi.nlm.nih.gov). Therefore, as previously discussed under previous nomenclature as Ugt1a11, mouse Ugt1a8 may represent a pseudogene (Zhang et al., 2004).

Mouse UGTs are widely expressed in a variety of other tissues (Table 2), some of which are involved in pharmacokinetics and others in endogenous steroid metabolism. Kidney is well known for urinary excretion. Several studies in humans, mice, and rats have identified the presence of UGT isozymes in kidney, specifically, human UGT1A3, 1A9, 2B4, 2B7, 2B10, 2B11, 2B15, and 2B17 and rat UGT1A1, 1A6, 1A7, and 2B8 (Emi et al., 1995; Beaulieu et al., 1996, 1998; Levesque et al., 1997, 1999; Grams et al., 2000; Turgeon et al., 2001; Shelby et al., 2003; Zhang et al., 2004). In the present study, six members of mouse UGTA and Ugt2a families were highly expressed in kidney. Both rats and mice express Ugt1a7 in kidney, suggesting species similarities among rodents.

In lung, six human UGT2B (4, 7, 10, 11, 15, and 17) isozymes have been detected (Turgeon et al., 2001), whereas three, UGT1A6, 1A7 (Emi et al., 1995), and 2B12 (Emi et al., 1995; Shelby et al., 2003), were detected in rat lung. In contrast, only one mouse isoform, Ugt1a6, was moderately expressed in lung. The presence of Ugt1a6 in lung may be attributed to the demand for glucuronidation of phenolic compounds from inspired air.

Mouse UGT expression was detected in steroid-sensitive tissues. Human UGT enzymes, particularly the UGT2B family, are expressed in steroid-sensitive tissues such as breast (mammary tissue), placenta, uterus, adrenal, prostate, and testis, and have been found to conjugate endogenous hormones (testosterone, dihydrotestosterone, androstenedione, estradiol, and some phenolic xenobiotics) (Turgeon et al., 2001). As reviewed by Turgeon et al. (2001), four human UGT2B isoforms (4, 10, 15, and 17) are expressed in testis, whereas UGT1A1, 1A6, and 1A7 have been detected in rat testis (Emi et al., 1995). Rat UGT1A6 is expressed in ovary, similar to what was observed here for mouse Ugt1a6 (Auyeung et al., 2001). The same laboratory also determined that the tissue-specific expression of rat UGT1A6 is dependent on basal activation of two separate 5′-proximal promoter regions, either P1 in spleen, lung, and ovary, or the P2 promoter in liver, kidney, and intestine (Auyeung et al., 2001). In mice, Ugt1a6, the phenol-conjugating isoform, was moderately expressed in testis and ovary, and only one Ugt2b mRNA, Ugt2b35, was expressed at low levels in mouse testis and ovary. Expression in testis and ovary may implicate mouse Ugt1a6 involvement in endogenous steroid conjugation. Mice may have a similar mechanism of controlling Ugt1a6 expression, as noted above for rats, based on its ubiquitous expression profile and the predicted alternative start site sequences (www.ensembl.org). In human placenta, six UGT2B members were expressed; however, in this study, only Ugt2b35 was barely detectable in mouse placenta, implicating differences in placental biotransformation between species (Collier et al., 2002). RNA from other steroid-sensitive tissues was not analyzed in this study, leaving in question the expression of UGTs in such tissues.

Mouse UGTs were detected in whole brain homogenates. Glucuronidation of endogenous neurotransmitters, and possibly toxins, occurs in the brain at the blood-brain barrier (King et al., 1999). In humans, UGT1A6 and 2B7 are expressed in adult cerebellum (King et al., 1999). Rats have minimal UGT expression in cerebellum and cerebral cortex (Shelby et al., 2003). Mouse Ugt1a7c has been reported in cerebrum and cerebellum (Zhang et al., 2004). In this study, low expression of Ugt2b35 was detected in homogenates of whole mouse brain.

Human and rat UGT2A1 genes were cloned from and are exclusively expressed in nasal epithelium (Jedlitschky et al., 1999; Heydel et al., 2001; Shelby et al., 2003). Tukey and Strassburg (2001) illustrated that human UGT2A1 underwent gene duplication to form UGT2A2, the tissue distribution of which differs from that of UGT2A1; UGT2A2 is expressed in small intestine and liver. Mouse Ugt2a1 and Ugt2a2 share high sequence homology and were detected together by a single bDNA probe set exclusively in nasal epithelia. However, Ugt2a3 is expressed in liver and small intestine, similar to human UGT2A2. Likewise, a novel subfamily of Ugt3a genes transcripts, named Ugt3a1 and 3a2 (Mackenzie et al., 2005), share significant sequence homology and were detected by a single probe set in liver and kidney in this study.

It is generally known that phase II conjugation reactions require high-energy cosubstrates. For glucuronidation, UDP-gpp and UDP-gdh metabolize glucose 1-phosphate to the high-energy cosubstrate donor UDP-GA. UDP-gpp was expressed in a wide range of mouse tissues, including liver, kidney, stomach, small and large intestine, heart, brain, and ovaries. This is not surprising because of the importance of UDP-glucose in general cellular intermediary metabolism, especially in liver. However, UDP-gdh was highly expressed in mouse GI tract, suggesting the importance of high-capacity glucuronidation of orally ingested xenobiotics in intestine, perhaps because of a decreased capacity to store UDP-GA in intestinal tissue versus liver.

The present study suggests potential gender differences in C57BL/6 mice. Female-predominant expression of Ugt1a1 in mouse liver correlates with gender differences in vivo bilirubin glucuronidation in Wistar rats (Muraca and Fevery, 1984), suggesting potential functional gender differences in mouse bilirubin metabolism. Further evidence indicates that rat UGT1A1 is repressed by testosterone in male rats and induced by progesterins in female rats, highlighting the roles of sex hormones in regulating UGT basal transcription (Muraca and Fevery, 1984). Additionally, rat UGT2B3 is predominant in males, in which testosterone exhibited an inductive effect, and estrogen slightly reduced basal expression (Strasser et al., 1997). In mouse liver, Ugt2b1 is male-predominant, whereas Ugt1a1 and 1a5 are female-predominant. In mouse kidney, Ugt2b5/37/38 is male-predominant and, conversely, Ugt1a2 and 1a10 are female-predominant. Whether these genes are repressed or induced by sex hormones, or alternative pathways such as growth hormone pattern secretion, has yet to be determined. Taken together, the results indicate that mouse UGTs are regulated by sex hormones.

Tissue-specific transcription factors control basal expression of UGTs. Mackenzie et al. (2003) reviewed tissue-specific transcription factors regulating human and rat UGTs. Those include hepatocyte nuclear factor 1α, CAAT-enhancer binding protein, octamer transcription factor 1, and putative pre-B cell homeobox-2, which can confer specific expression of distinct UGT isoforms in liver and intestine. In addition, mice lacking C/EBP have deficiencies in hepatic expression of Ugt1a1 and Ugt2b (Lee et al., 1997). These factors.
undoubtedly contribute to tissue-specific UGT expression patterns observed in mice.

In the present study, 11 mouse UGTs were expressed in liver, 7 in GI tract, and 6 in kidney. Gender-specific gene expression was observed in liver and kidney. Similarities in expression patterns between species (humans, rats, and mice) were observed in tissues and between orthologous isozymes. Taken together, these data can potentially be used to determine tissue-specific-dependent glucuronidation based upon substrate specificity and tissue localization of individual UGT isoforms. Additional insights into gender-dependent differences in drug metabolism in vivo are shown here as well.

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References


