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Cloning, Expression, and Ontogeny of Mouse Organic Anion-Transporting Polypeptide-5, a Kidney-Specific Organic Anion Transporter[1]

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The full-length coding sequence of mouse organic anion-transporting polypeptide (designated mouse Oatp-5) has been cloned from mouse kidney cDNA library. Analysis of the 5′-untranslated region (5′-UTR) of Oatp-5 cDNA through capsite cloning reveals two possible transcription start sites that are 4-bp apart. The 3′-untranslated region (3′-UTR) of Oatp-5 cDNA contains an early polyadenylation signal, indicating the possibility that mRNAs with different 3′-UTR lengths may coexist. Deduced amino acid sequence of mouse Oatp-5 protein contains 670 amino acids and has 10 putative transmembrane domains, multiple potential glycosylation and phosphorylation sites. Tissue-specific expression studies indicate that mouse Oatp-5 is expressed only in kidney. Studies on the developmental expression reveal that there is no significant expression of Oatp-5 mRNA in mouse kidney for at least 3 weeks after birth, and adult levels of Oatp-5 mRNA expression are attained more than 6 weeks after birth. Phylogenetic analysis reveals that mouse Oatp-5 is an ortholog of rat Oatp-5.

Renal functions, such as urine concentrating capacity, acidification ability and glucose transport ability, are poorly developed at birth (1, 2). Some of these functions mature rapidly during the perinatal period. For example, the glomerular filtration rate (GFR) increases twofold during the first two weeks of life (3). Because kidney is the organ central to the maintenance of homeostasis, the perinatal maturation of renal functions is important for the body to adapt to extrauterine life.

Tubular secretion of organic anions is one of the essential functions of the kidney that has been extensively characterized using *para*-aminohippuric acid (PAH) as a prototypical substrate (4, 5). A variety of xenobiotics, toxins, endogenous compounds and their metabolites are classified as organic anions, and many of these organic anions are eliminated via carrier-mediated pathway(s) in the proximal tubules. A number of drugs, such as β-lactam antibiotics, diuretics, non-steroidal anti-inflammatory drugs (NSAIDs) and several anti-viral drugs are also classified as organic anions. Therefore, renal organic anion transport pathway plays a key role in the pharmacokinetics of these drugs as well as other xenobiotics.

Using molecular cloning techniques, a variety of cDNAs encoding transporters and channels in the renal tubules have been cloned, and the ontogenic expression of several of these transporters has been reported (6). Sequence analysis of these transporters indicates that they belong to distinct families. For example, the organic anion transporting polypeptide (Oatp) family, organic anion transporter (OAT) family and kidney-specific organic anion transporter (OAT-K1 and OAT-K2) family. While OAT-K1 and OAT-K2, cloned from rat kidney, are expressed only in kidney (7, 8), members of the OAT as well as Oatp families are expressed in multiple tissues. Members within a family often share amino acid sequence identity of greater than 70%, members from different families may share identity less than 40%.

All members of the organic anion transporting polypeptide (Oatp) family reported so far are expressed in multiple tissues. For example, rat Oatp-1 and Oatp-2 are expressed in liver, kidney and brain (9, 10),

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and Oatp-3 is expressed in brain and liver (11). Rat Oatp-4 reported recently (12), is actually same as rat lst (liver-specific organic anion transporter) (13). Sequence as well as phylogenetic analysis revealed that rat Oatp-4 does not bear any significant similarity to other members of the Oatp family but it is an ortholog of human LST-1 and mouse lst-1 (14, 15). In this communication, we report the cloning of a new member of the Oatp family from mouse kidney cDNA library (Oatp-5). We also report the tissue-specific expression and ontogeny of mouse Oatp-5, and its relationship to other members of the Oatp family as well as to other organic anion transporters.

MATERIALS AND METHODS

Materials. Restriction enzymes were obtained from Gibco-BRL (Gaithersburg, MD). Terminal transferase was purchased from Roche Molecular (Indianapolis, IN). [α-^{32}P]dATP and Ready-To-Go DNA ligase were from Amersham Pharmacia Biotech (Piscataway, NJ). Zeta probe membrane was obtained from Bio-Rad (Hercules, CA).

cDNA cloning. cDNA cloning of mouse Oatp-5 was performed by 5′- and 3′-RACE (Rapid Amplification of cDNA Ends) reactions using BALB/c mouse kidney-derived adaptor-ligated Marathon-Ready cDNA library (Clontech, Palo Alto, CA). 5′-RACE reactions using the primer 5′-GCTAGGCCATTGTTCCCACACACTGGATCC-3′ yielded a ~1.5 kb fragment, and 3′-RACE reactions using the primer 5′-AAAGAGGAGAAGCACAGAGACAAGGCCAAG-3′ yielded a ~1.7 kb fragment. These fragments had a 500-bp overlap. A touchdown PCR was performed for RACE reactions with the following conditions: 1 cycle of 94°C × 30 s; 6 cycles of 94°C × 5 s, 72°C × 4 min; 6 cycles of 94°C × 5 s, 70°C × 4 min; 26 cycles of 94°C × 5 s, 68°C × 4 min; and a final extension of 72°C × 7 min. The RACE products were subcloned into pT-Adv TA cloning vector (Clontech, Palo Alto, CA), and sequenced with an ABI Prism 377 DNA sequencer (Perkin Elmer, Foster, CA). To confirm the accuracy of the sequence, a total of three independent reactions were performed. Two different types of clones were obtained, both had the full-length coding sequence, but different lengths of the 3′-UTR. Sequencing of the cDNAs was done by primer walking, and both strands were sequenced. Synthesis of all oligonucleotide primers and sequencing were carried out by the Biotech Support Facility, University of Kansas Medical Center.

Capsite cloning. Capsite cloning was performed using the First Choice RLM RACE kit (Ambion, TX). The technique is based on RNA ligase-mediated and capped mRNA-dependent rapid amplification of cDNA ends (RLM-RACE). Briefly, about 500 ng poly(A)$^+$-enriched RNA was treated with calf intestinal phosphatase (CIP) to remove the 5′-PO_4 group from the non-capped (5′-end truncated) mRNAs. CIP leaves the 5′-m_7-GTP cap of the full-length mRNAs intact. 5′-dephosphorylated RNA was then treated with tobacco acid pyrophosphatase (TAP) which specifically removes the cap structure leaving a 5′-PO_4 intact. These full-length decapped mRNAs were then ligated at the 5′-end by RNA ligase, to an RNA oligo adapter provided by the manufacturer. First strand cDNA synthesis was done by reverse transcription of this adapter-mRNA template, using random primers. 5′-RACE was performed using a gene specific reverse primer (Reverse GSP) mentioned above for cloning the mouse Oatp-5, and an outer adapter primer (forward primer) provided by the manufacturer. This reaction was used for a nested PCR using a nested gene specific reverse primer (5′-CACACACTCTGCTGGGTCTTGTGTTGGCTTT-3′) and an inner adapter primer (forward primer) provided by the manufacturer. The amplification product (a single band) was subcloned in pT-Adv TA cloning vector (Clontech, Palo Alto, CA), and a number of clones from independent reactions were sequenced to determine the entire 5′-UTR including the capsite.

TABLE 1

Similarity in the Nucleotide Sequence and Protein Sequence (in Parentheses) of the Coding Region of Mouse Oatp-5 and Other Transporters

	moatp-5		moatp-5
moatp-1	83.6 (73.6)	roatp-1	82.2 (71.6)
moatp-2	85.0 (75.4)	roatp-2	84.1 (76.0)
mlst-1	50.9 (39.3)	roatp-3	84.6 (75.7)
		roatp-5	87.3 (80.4)
hOATP-1	75.7 (65.7)	rlst-1a	49.8 (40.1)
hOATP-2	50.9 (41.1)	roat-k1	83.5 (71.0)
hOATP-8	51.5 (43.7)	roat-k2	81.8 (73.3)
hPGT	47.5 (34.0)	rPGT	46.7 (34.0)

Note. The prefixes m, r, and h in the names of the transporters stand for mouse, rat, and human, respectively. Values are percentages.

Northern blot analysis of rat lst-1 mRNA. Total RNA was isolated from pooled liver, kidney, brain, heart, lungs and intestine from 10-week-old BALB/c male mice (3 mice for each tissue), using Trizol reagent following manufacturer's direction. The rationale for using 10-week-old mice is that their age matches the age of the BALB/c mice (9–11 weeks old) from which the Marathon-Ready cDNA library was prepared. Poly(A)$^+$ mRNA from each tissue was purified from total RNA using poly(A)$^+$ pure mRNA isolation kit from Ambion (Austin, TX). Poly(A)$^+$-enriched RNA was electrophoretically resolved in 1% formaldehyde-agarose gel, capillary-blotted overnight onto Zeta-probe nylon membrane, UV cross-linked and hybridized at 46°C overnight in 1× hybridization solution (Sigma, St. Louis, MO) containing 25% formamide. For the tissue distribution study, 3 μg, and for the ontogeny study 6 μg poly(A)$^+$-enriched RNA was used in each lane. Antisense oligonucleotide probes were 3′-end labeled using terminal transferase and the average specific activity was about 5×10^8 cpm/μg DNA. More than one oligonucleotide probe was used to increase the sensitivity of detection. The probe sequences were as follows: Probe 1, 5′-CCCAGAGTTCAGATCACTCCAAACTTGTCC-3′; Probe 2, 5′-AGCAGCTGAGGAAGCAGCTGTCCTCAGTGC-3′; Probe 3, 5′-AAATTCAATGAAATTAAGTGAAATGAAATC-3′. These antisense oligonucleotide probes were designed after aligning the cDNA sequences of Oatp-5 with other organic anion transporters to choose a region that is unique to Oatp-5.

Computational biology. DNA sequence analysis, alignments, contig assembly, amino acid sequence prediction and construction of phylogenetic tree were done by a combination of DNAsis, Autoassembler and Clutal W software. Prediction of putative membrane topology was done with the aid of hidden Markov model for predicting transmembrane helices (http://www.cbs.dtu.dk/services/TMHMM-1.0).

RESULTS AND DISCUSSION

Mouse Oatp-5 Belongs to the Organic Anion Transporting Polypeptide (Oatp) Family and It Bears the Highest Sequence Identity with Rat Oatp-5

The open reading frame (ORF) of mouse organic anion transporting polypeptide-5 (Oatp-5) cDNA encodes a putative protein of 670 amino acids with a predicted molecular mass of approximately 74 kDa. Table 1 shows the percent sequence identity of both nucleotides and amino acids (within parenthesis) between mouse Oatp-5 and several other organic anion

transporters in mice, rats and humans. Mouse Oatp-5 shows greater than 80% sequence identity at the nucleotide level, and greater than 70% sequence identity at the amino acid level, with other members of the Oatp family of both mice and rats (9–11, 16). Among the human OATPs, mouse Oatp-5 shows the greatest sequence identity with human OATP-1 (17). Mouse Oatp-5 bears the highest sequence identity with rat Oatp-5 (87.3% at the nucleotide level and 80.4% at the amino acid level). Rat Oatp-5 has been recently reported in GenBank only (Accession No. AF053317) (18).

Figure 1a shows the amino acid sequence comparison between mouse Oatp-5 and rat Oatp-5. There are several regions common to mouse Oatp-5 and rat Oatp-5, that have highly conserved amino acid sequence. The two longest such regions are: aa 84 through aa 203, and aa 396 through aa 553. In these regions, the amino acid sequence identity is approximately 90% or greater. However, there are regions that are significantly different between mouse Oatp-5 and rat Oatp-5 (amino acid sequence identity around 50% or less). Two such regions are: aa 275 through aa 299, and aa 623 through aa 659. Other regions of dissimilarity are even smaller, usually 10–12 amino acid long. The extent of divergence in these regions in the face of a high overall conservation of amino acid sequence between mouse Oatp-5 and rat Oatp-5 is suggestive of their lack of contribution towards the functional morphology of these proteins.

Capsite Cloning Reveals That the Transcription Initiation Site of Mouse Oatp-5 mRNA Starts with an Adenine (A) and There Are Two Putative Transcription Initiation Sites in Mouse Oatp-5 Gene

In order to recover more of the 5′-UTR sequence, 5′-RACE was performed using different primers. Sequencing of the products revealed that these additional 5′-RACE reactions did not yield any additional 5′-UTR sequence compared to that obtained in the initial 5′-RACE reactions. Therefore, capsite cloning was performed in order to determine if the 5′-UTR already obtained, was complete or any part of it was missing. Because the 5′-end-truncated (non-capped) mRNAs are dephosphorylated leaving only capped mRNAs for adapter ligation and subsequent amplification, it is a highly reliable technique for obtaining full-length capped mRNAs. The result of capsite cloning shows that the transcription initiation site (capsite) of mouse Oatp-5 mRNA starts with an adenine (A), and the entire 5′-UTR of mouse Oatp-5 is 178-bp long (Fig. 1b). Interestingly, two different types of clones were obtained. Compared to the 5′-UTR obtained by initial 5′-RACE, the longer clones had 19 additional bp of the 5′-UTR (starts as 5′-ATCC. . .), and the shorter clones had 15 additional bp of the 5′-UTR (starts as 5′-ATTC. . .). Hence, the shorter clones lack the first four bases of the longer clones (Fig. 1b). Thus, it appears that mouse Oatp-5 gene transcription can be initiated at two different sites that are 4-bp apart, as shown by arrows in Fig. 1b. A similar situation was reported for the trnN2 gene, where two transcription initiation sites were identified that are 7-bp apart (19).

During the cloning of Oatp-5, cDNA clones of two different lengths were obtained. The shorter cDNA had a polyadenylation signal (AATAAA) 386 bp downstream from the stop codon, and it was truncated and polyadenylated 12 bp downstream from this polyadenylation signal. For the longer clones, an additional 216 bp region was obtained beyond the polyadenylation signal, but no typical polyadenylation signal was found in this additional region. The presence of this early polyadenylation signals in the 3′-untranslated region (UTR) of mouse Oatp-5 indicates the possibility that functional transcripts with different 3′-UTR lengths may co-exist. The phenomenon of alternative polyadenylation by using different polyadenylation signals in the 3′-UTR, has been reported for a number of transcripts, and it is usually associated with differential stability of the transcript (20). Alternative polyadenylation can also be observed in transcripts expressed in different organs where their half-lives may be different. In the case of mouse Oatp-5, the significance of alternatively polyadenylated mRNAs co-existing in the same organ (kidney), remains to be explained.

Mouse Oatp-5 Protein Contains 10 Putative Transmembrane Domains and Several Potential Amino Acid Modification Sites

Prediction of possible topology of mouse Oatp-5 (and also rat Oatp-5) was performed by hidden Markov model for predicting transmembrane helices (TMHMM program), and the results are presented in Fig. 2a. Mouse Oatp-5 has 10 putative transmembrane domains (TMDs). This is similar to rat lst-1c (13) which also has 10 TMDs, but unlike most other Na^+-independent organic anion transporters which have 12 TMDs. Three out of ten TMDs, i.e., TMD# 3, 4 and 10 are each 19 amino acids long while the rest are 23 amino acids long. The protein appears to contain one large and one moderate extracellular loop domain but only one large intracellular loop domain. The intracellular C-terminal domain of Oatp-5 is very long and hydrophilic and it contains several putative modification sites.

Analysis of the putative amino acid modification sites (http://maple.bioc.columbia.edu/predictprotein/ and http://www.cbs.dtu.dk/services/NetOGly) indicates that mouse Oatp-5 protein possesses 5 putative protein-kinase C (PKC) phosphorylation sites: Thr 351, 580, 645, Ser 596 and 657. Except Thr 351, all other

a

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moatp-5         MGEPGKRVGIHRVRCFAKIKVFLLALIWAYISKILSGVYMSTMLTQLERQFNISTSIVGL
roatp-5         MGEPEKRAGTHGIRCFAKIKVFLLALTWAYASKALSATYMNSMLTQIERRFNISTSIVGL
                **** **.* * :************** *** ** **..**.:****:**:**********

moatp-5         INGSFEMGNLLVIVFVSYFGTKLHRPIMIGVGCAVMGLGCFIISLPHFLMGRYEYETTIS
roatp-5         INGSFEVGNLLLIIFVSYFGRKRHRPIMIGIGCAVMGLGCFIISLPHFLMGRYEYETTIS
                ******:****:*:****** * *******:*****************************

moatp-5         PTSNLSSNSFLCVENRSQTLKPTQDPAECVKEIKSLMWIYVLVGNIIRGIGETPIMPLGI
roatp-5         PTSNLSSNSFLCMENRTQTLKPTQDPAECVKEMKSLMWIYVLVGNIIRGIGETPIMPLGI
                ************:***:***************:**************************

moatp-5         SYIEDFAKSENSPLYIGILEVGKMIGPILGYLMGPFCANIYVDTGSVNTDDLTITPTDTR
roatp-5         SYIEDFAKSENSPFYIGILEVGKITGPIAAIWLGSFCATIYVDMGSVNTDDLTITPTDTR
                *************:*********: *** .  :*.***.**** ****************

moatp-5         WVGAWWIGFLVCAGVNVLTSIPFFFFPKTLPKEGLQDNGDGTENAKEEKHRDKAKEENQG
roatp-5         CVGAWWIGFLVCAGLNILISIPFFFFPKTFPKEGPEDMANETKNDEGDKHREKAKEEKRG
                 *************:*:* **********:**** :* .: *:* : :***:*****::*

moatp-5         IIKEFFLMMKNLFCNPIYMLCVLTSVLQVNGVANIVIYKPKYLEHHFGISTAKAVFLIGL
roatp-5         ITKDFFLFMKSLSCNPIYMLCVLTSVLQVNGFVSIFTFKPKYLEHHYGKSSSEAIFLMGL
                * *:***:**.* ******************...*. :********:* *:::*:**:**

moatp-5         YTTPSVSAGYLISGFIMKKLKITLKKAAIIALCLFMSECLLSLCNFMLTCDTTPIAGLTT
roatp-5         YTLPSVCVGYLISGFIMKKFKITLKKAAFISYCLGMSECLLSLCNFMLTCDNVPIAGLTT
                ** ***..***********:********:*: ** ****************..*******

moatp-5         SYEGIQQSFDMENKFLSDCNTRCNCLTKTWDPVCGNNGLAYMSPCLAGCEKSVGTGANMV
roatp-5         SYEGIQQSFDMENTVLADCNTRCSCLTKTWDPVCGDNGLAYITPCLAGCEKSVGSGINMV
                *************..*:******.***********:*****::***********:* ***

moatp-5         FQNCSCIRSSGNSSAVLGLCKKGPDCANKLQYFLIITVFCCFFYSLATIPGYMVFLRCMK
roatp-5         LQDCSCIQSSGNSSAVLGLCNKGPDCANKLQYFLIITVFCSFFYSLSLIPGYMIFLRCMK
                :*:****:************:******************.*****: *****:******

moatp-5         SEEKSLGIGLQAFFMRLFAGIPAPIYFGALIDRTCLHWGTLKCGEPGACRTYEVSSFRRL
roatp-5         SEEKSLGIGLQAFCMRILGGILAPIYFGVLIDRTCLHWGTQKCGEPGACRTYEINSFRSI
                ************* **::.** ******.*********** ************:.*** :

moatp-5         YLGLPAALRGSIILPSFFILRLIRKLQIPGDTDSSEIELAETKPTEKESECTDMHKSSKV
roatp-5         YLGLPAALRGSSYLPAFFILRLMRKFQFPGDINSPVTDHVEMMLTEKESEHTDVHRSPQV
                ***********  **:******:**:*:*** :*.  : .*   ****** **:*:*.:*

moatp-5         ENDGELKTKL
roatp-5         ENDGELKTKL
                **********
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b

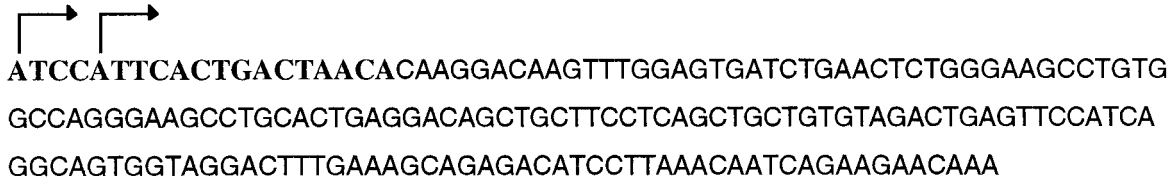

FIG. 1. (a) Comparison of the amino acid sequence of mouse Oatp-5 and rat Oatp-5. Asterisks indicate the conserved amino acids between the two sequences, and (b) the entire 5′-UTR of mouse Oatp-5 mRNA. Bases in bold are the ones obtained through capsite cloning. The two predicted transcription start sites (capsites) are indicated by arrows. Both start with an adenine (A).

sites are in the intracellular C-terminal domain. In addition, there are 2 putative protein-kinase A (PKA) phosphorylation sites: Ser 490 and 611, the latter one being in the intracellular C-terminal domain. Mouse Oatp-5 protein also has several potential glycosylation sites, 3 potential O-linked glycosylation sites: Thr 118,

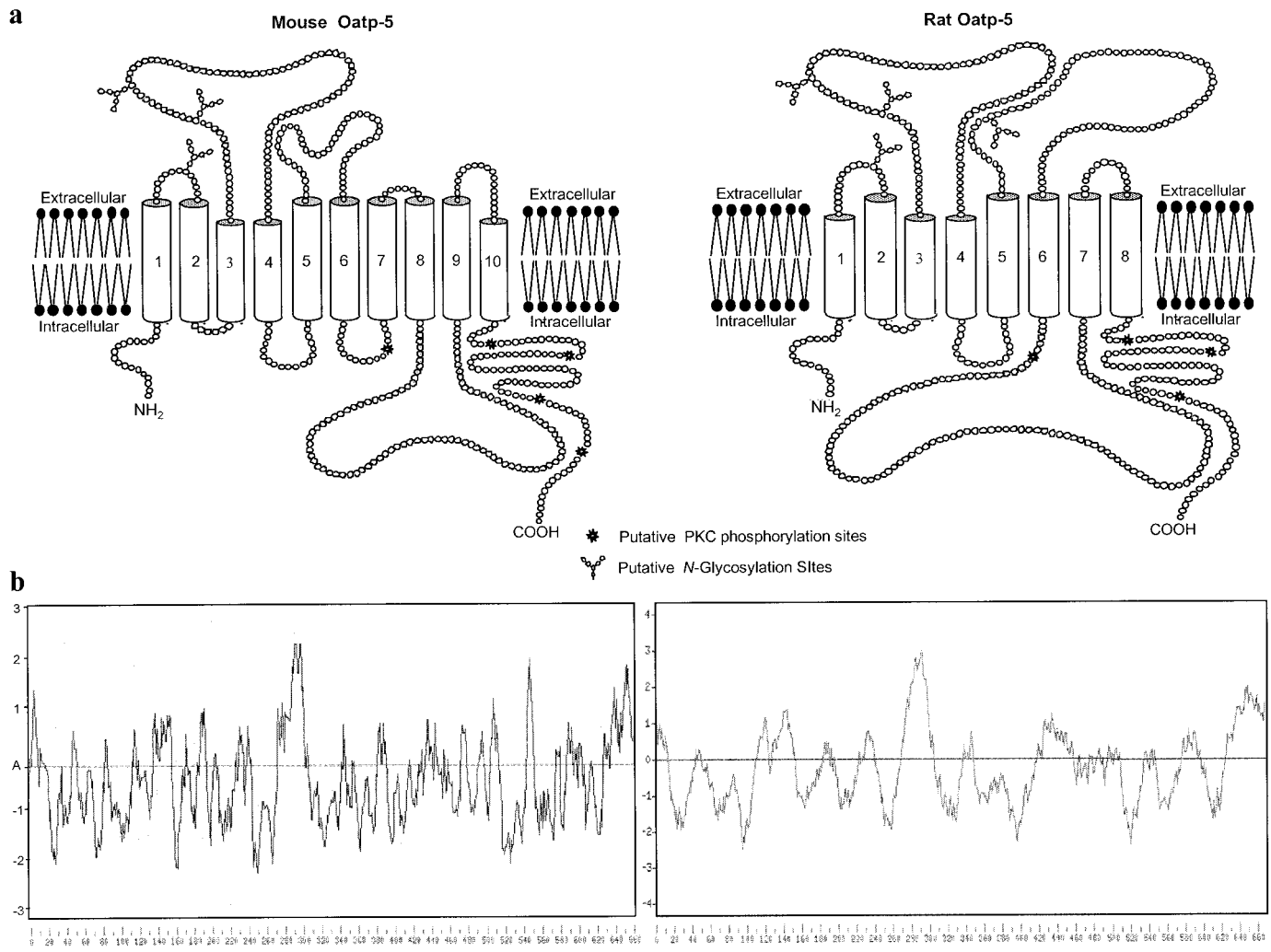

FIG. 2. (a) Topology of mouse Oatp-5 and rat Oatp-5 proteins. The transmembrane domains are indicated by cylinders. Each circle in the extracellular and intracellular loop represents one amino acid. Putative N-glycosylation and PKC-phosphorylation sites are also shown, and (b) Hopps and Wood's antigenicity plot (left), and Kyte and Doolittle hydrophilicity plot (right) of mouse Oatp-5 protein.

122 and 143, and 3 potential N-linked glycosylation sites: Asn 52, 124 and 135. Except Asn 52, all other sites are restricted to the large extracellular loop domain (Fig. 2a).

In contrast to mouse Oatp-5, rat Oatp-5 contains 8 putative TMDs, and three of them (TMD 1, 3 and 4) are 19 amino acids long while the rest are 23 amino acids long. The first two extracellular domains, and first three intracellular domains as well as the last C-terminal domain of both mouse and rat Oatp-5 are almost identical, including the putative N-glycosylation and PKC-phosphorylation sites. However, after TMD 5, rat Oatp-5 appears to have an additional large extracellular domain and a very large intracellular domain, compared to mouse Oatp-5 (Fig. 2a).

Hydrophobicity/hydrophilicity as well as antigenicity analysis (21, 22) of mouse Oatp-5 protein reveals that the two strongest antigenic and hydrophilic sections of the protein span approximately between aa 270 and 300, and aa 630 and 660. Interestingly, these are the regions where the amino acid sequence between mouse Oatp-5 and rat Oatp-5 are the most divergent (Fig. 2b).

Mouse Oatp-5 Is Constitutively Expressed in Kidney

Constitutive expression levels of mouse Oatp-5 in six different tissues were determined by northern-blot analysis as described earlier (13) (Fig. 3). Approximately 3 μg of poly(A)$^+$-enriched RNA was used for each tissue sample. Oatp-5 was found to express only in kidney. Determination of relative mobility shows that the size of Oatp-5 mRNA is approximately 3.2 kb. In a separate experiment using branched DNA (bDNA) signal amplification technology (23), it was found that rat Oatp-5 (mouse Oatp-5 ortholog) is also expressed

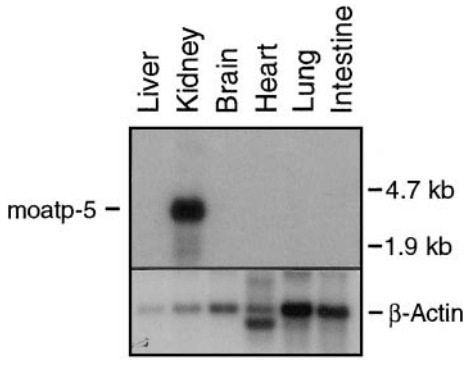

FIG. 3. Expression of Oatp-5 in different tissues in mouse. Upper panel shows Northern blot done with Oatp-5-specific antisense oligoprobes (see Materials and Methods), and lower panel shows antisense β-actin probe used as control. Each lane contains 3 μg of poly(A)$^+$ enriched RNA. The expression of Oatp-5 is kidney-specific.

only in kidney out of over 20 tissues tested (Hartley, Cherrington, and Klaassen; unpublished data).

Mouse Oatp-5 Expression Increases with Age

Postnatal expression of Oatp-5 was studied in 5-, 10-, 20-, 40-, and 60-day-old mice. For every sample, 6 μg of poly(A)$^+$-enriched RNA was used to ensure that even very low level expression would be detected. As evident from Fig. 4, the expression of Oatp-5 mRNA was detected from day 40 onward, and there appears to be a progressive increase in the expression with age at this time. The adult level was detected in the kidney of day 60 mice, indicating that the adult level is reached sometimes after 6 weeks. Interestingly, no significant expression of Oatp-5 mRNA was observed at least up to 3 weeks after birth. In a similar experiment with rat Oatp-5 (mouse Oatp-5 ortholog), using bDNA signal amplification technology, a significant increase in the expression of rat Oatp-5 was observed after day 40 (Hartley, Cherrington, and Klaassen; unpublished data).

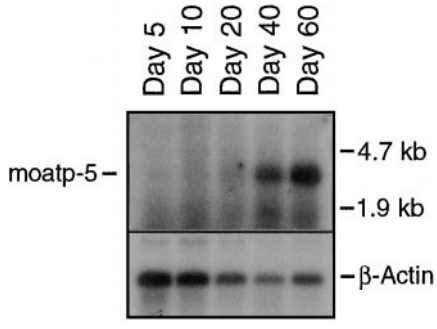

FIG. 4. Postnatal expression of Oatp-5 in mouse. Upper panel shows Northern blot done with Oatp-5-specific antisense oligoprobes (see Materials and Methods), and lower panel shows antisense β-actin probe used as control. Each lane contains 6 μg of poly(A)$^+$-enriched RNA. The expression of Oatp-5 increased with age, and the adult level was reached after day 40.

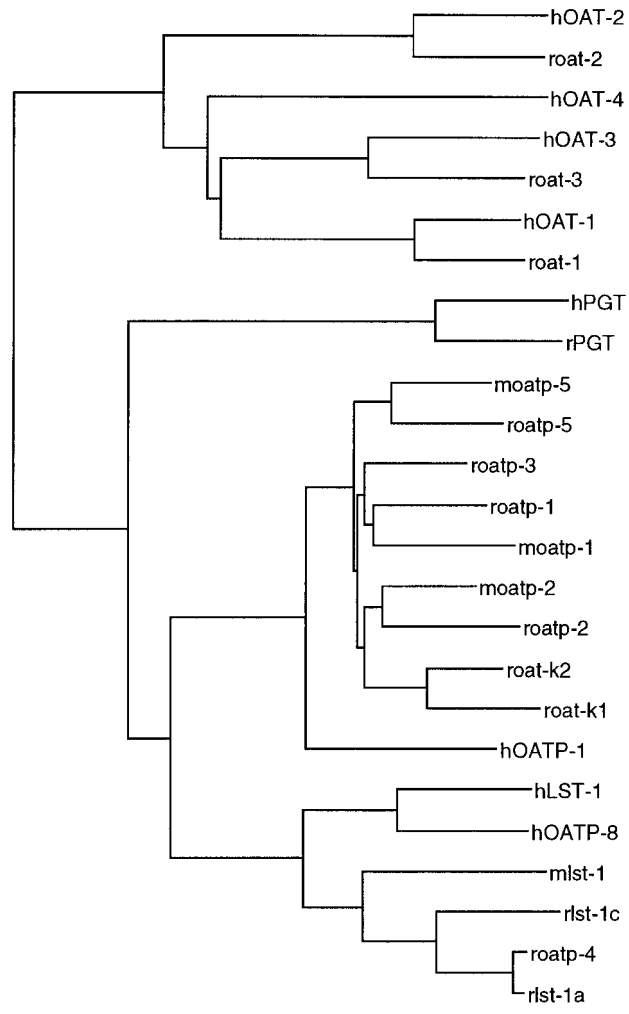

FIG. 5. Phylogenetic analysis reveals that mouse Oatp-5 is an ortholog of rat Oatp-5, and they diverged from the rest of the Oatps from a common root.

Mouse Oatp-5 is an Ortholog of Rat Oatp-5

In an attempt to understand the evolutionary relationship/divergence of the various families of organic anion transporters across species, such as, Oatps, PGTs, lst-1, OATs as well as kidney-specific organic anion transporters (OAT-K1, OAT-K2), nucleotide sequences of the coding regions of the cDNAs from mouse, rat, and human were all aligned and a phylogenetic tree was constructed (Fig. 5). There were four major clusters: (1) rat and human OATs; (2) rat and mouse Oatps, OAT-K1 and OAT-K2; (3) rat and human PGT; and (4) rat and mouse lsts, human LST-1 and OATP-8. Interestingly, human OATP-1 clustered with rat and mouse Oatps. Phylogenetic analysis thus strongly suggests that mouse Oatp-5 and rat Oatp-5 are orthologs and they diverged from the rest of the oatps from a common root.

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