Transport of ^{99m}Tc-MAG3 via Rat Renal Organic Anion Transporter 1

Naoto Shikano, MS[1]; Yoshikatsu Kanai, MD, PhD[2]; Keiichi Kawai, PhD[3]; Nobuyoshi Ishikawa, MD, PhD[1]; and Hitoshi Endou, MD, PhD[2]

[1]Department of Radiological Sciences, Ibaraki Prefectural University of Health Sciences, Ibaraki, Japan; [2]Department of Pharmacology and Toxicology, Kyorin University School of Medicine, Tokyo, Japan; and [3]School of Health Sciences, Faculty of Medicine, Kanazawa University, Kanazawa, Japan

Recently, complementary DNA (cDNA) encoding a *p*-aminohippurate (PAH) transporter designated rat organic anion transporter 1 (OAT1) was isolated. OAT1, a multispecific organic anion transporter at the basolateral membrane, is exclusively expressed in the middle segment of the proximal tubule in the rat kidney. It has been proposed that OAT1 is indirectly involved in PAH uptake via the Na^+ dicarboxylate cotransporter. In this study, in molecular biologic experiments using OAT1-expressing *Xenopus laevis* oocytes, we obtained evidence that ^{99m}Tc-mercaptoacetylglycylglycylglycine (MAG3) is transported via OAT1. **Methods:** Capped OAT1 complementary RNA (cRNA) was synthesized from library plasmid cDNA linearized with *Bam*HI using in vitro transcription. Defolliculated oocytes were injected with 10 ng of OAT1 cRNA. Two to 3 d after injection, uptake of ^{99m}Tc-MAG3 was measured using ND96 solution containing 18.5 kBq of ^{99m}Tc-MAG3. Before the uptake experiments, OAT1-expressing oocytes were preincubated for 2 h with 1 mmol/L glutarate (a dicarboxylate), to generate an outwardly directed glutarate gradient. Then, after incubation for 60 min at room temperature, radioactivity of oocytes was determined. For the inhibition experiments, uptake was assessed in the absence or presence of inhibitor: 2 mmol/L of PAH, *o*-iodohippurate (OIH), probenecid, 3,5-diiodo-4-pyridone-*N*-acetate (iodopyracet), furosemide, ethacrynic acid, glucoheptonate, maleic acid, L-Tyr, or tetraethylammonium (TEA) or 0.1 mmol/L of 2,4-dinitrophenol (DNP). **Results:** Na^+ had a significant effect on ^{99m}Tc-MAG3 uptake ($P < 0.05$). Accumulated glutarate stimulated simultaneous ^{99m}Tc-MAG3 uptake and glutarate excretion ($P < 0.001$). The following compounds significantly inhibited ^{99m}Tc-MAG3 uptake: PAH, 8.5% ± 16.2% of ^{99m}Tc-MAG3 uptake in the absence of an inhibitor; OIH, 26.4% ± 21.7%; probenecid, 29.1% ± 12.4%; iodopyracet, 15.8% ± 7.9%; furosemide, 30.5% ± 15.7%; ethacrynic acid, 21.6% ± 10.6%; glucoheptonate, 35.6% ± 22.6%; and maleic acid, 60.1% ± 18.7%. ^{99m}Tc-MAG3 accumulation in *Xenopus laevis* oocytes was not significantly inhibited by TEA, L-Tyr, or DNP. **Conclusion:** The following substances had a *cis*-inhibitory effect on ^{99m}Tc-MAG3 transport: PAH, OIH, probenecid, iodopyracet, furosemide, ethacrynic acid, and glucoheptonate. Glutarate had a *trans*-stimulative effect on ^{99m}Tc-MAG3 transport. ^{99m}Tc-MAG3 acts as a substrate of OAT1, an organic anion/dicarboxylate exchanger.

Key Words: ^{99m}Tc-MAG3; organic anion transporter 1; *Xenopus laevis* oocyte; membrane transport; renal function

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The organic anion ^{99m}Tc-mercaptoacetylglycylglycylglycylglycine (MAG3) was first used to replace ^{131}I-*o*-iodohippurate (OIH) in renal function studies in 1986 (*1*). Compared with ^{99m}Tc-MAG3, ^{131}I-OIH gives poor spatial resolution because the permissible injection dose is limited and its photon energy requires the use of coarse-resolution collimators (*2*). Also, ^{99m}Tc-MAG3 has been shown to have biologic properties similar to those of ^{131}I-OIH in animals and humans and to be a superior single-photon emitter in scintigraphy (*1*,*3*). Renal scintigraphy with ^{99m}Tc-MAG3 can provide excellent image quality, even in patients with severely decreased renal function (*4*).

Organic anion transporters have been defined as transporters of organic compounds that are inhibited by an excess of *p*-aminohippurate (PAH) (*5*). They are important transporters, because many clinically important drugs and toxic compounds are organic anions, and these compounds are eliminated via the organic anion transport system (*5*). Generally, transepithelial secretion of organic anions occurs via accumulative transport from blood into proximal tubule cells across the basolateral membrane (*6*). Organic anions such as ^{99m}Tc-MAG3 are excreted through the proximal tubular cells of the nephron in a PAH-competitive manner (*4*).

Recently, Sweet et al. and Sekine et al. independently isolated complementary DNA (cDNA) encoding a PAH transporter designated rat organic anion transporter 1 (OAT1; Fig. 1) (*6*,*7*). OAT1, a multispecific organic anion transporter at the basolateral membrane, is exclusively expressed in the middle segment of the proximal tubule in the rat kidney (*6*). To date, 4 isoforms of mammalian OAT have been identified (*6–10*). It is unclear which transporter is

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For correspondence or reprints contact: Naoto Shikano, MS, Department of Radiological Sciences, Ibaraki Prefectural University of Health Sciences, 4669-2 Ami, Ami-machi, Inashiki-gun, Ibaraki 300-0394, Japan.

E-mail: sikano@ipu.ac.jp

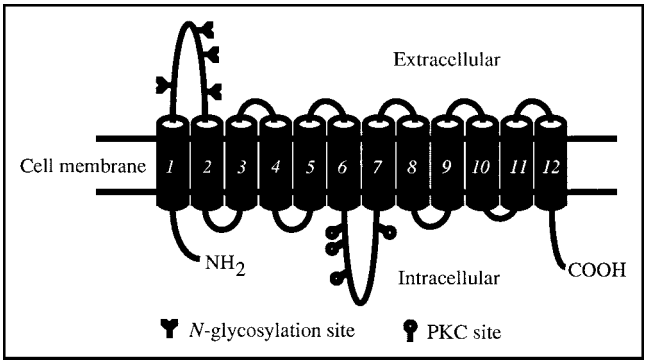

FIGURE 1. Putative membrane topology of OAT1. OAT1 comprises 551 amino acids and has 12 putative membrane-spanning domains. Four *N*-glycosylation sites and 4 protein kinase C–dependent phosphorylation sites (PKC sites) have been predicted in first and sixth hydrophilic loops, respectively.

involved in transport of ^{99m}Tc-MAG3, which is widely used as the imaging agent of choice for renal scintigraphy (*4*). In the present study, molecular biologic experiments showed that ^{99m}Tc-MAG3 is transported via OAT1 expressed by *Xenopus laevis* oocytes.

MATERIALS AND METHODS

Isolation of *Xenopus laevis* Oocytes

The ethical committees of all relevant universities approved the animal experiments. All chemicals used were reagent grade (Aldrich Chemical Co. and Kanto Chemical Co., Inc.). Collagenase was purchased from Boehringer Mannheim. Adult female *Xenopus laevis* (Japan SLC Inc.) were anesthetized by hypothermia and decapitated. Ovaries were then removed and stored in OR2 solution. Stage V and VI oocytes were manually dissected from the ovary. *Xenopus laevis* oocyte follicles were removed by digestion in OR2 solution (pH 7.5) containing 82.5 mmol/L NaCl, 2 mmol/L KCl, 1 mmol/L $MgCl_2$, 5 mmol/L 2-hydroxyethylpiperazine-*N*′-2-ethanesulfonic acid (HEPES), and 1.5 mg/mL collagenase, for 30–40 min at room temperature. In oocyte experiments, 6–22 oocytes were used.

Complementary RNA (cRNA) Synthesis and Oocyte Injection

Capped OAT1 cRNA (GenBank/EBI Data Bank accession no. AB004559) was synthesized in vitro using T7 RNA polymerase (Stratagene) and plasmid DNA linearized with *Bam*HI (Invitrogen), as described elsewhere (*6*). Defolliculated oocytes were injected with 10 ng of capped cRNA and then incubated in modified Bath's solution (pH 7.4) containing 88 mmol/L NaCl, 1 mmol/L KCl, 0.33 mmol/L $Ca(NO_3)_2{\cdot}4H_2O$, 0.4 mmol/L $CaCl_2{\cdot}2H_2O$, 0.8 mmol/L $MgSO_4{\cdot}7H_2O$, 2.4 mmol/L $NaHCO_3$, 10 mmol/L HEPES, and 150 mg/mL gentamicin, at 18°C for 2–3 d.

Uptake Assays

^{99m}Tc-MAG3 was obtained from Nihon Medi-Physics Co. and Daiichi Radioisotope Laboratories Ltd. and was purified by high-performance liquid chromatography under the conditions described by Shattuck et al. (*11*).

The uptake experiments were performed using ND96 solution (pH 7.4) containing 96 mmol/L NaCl, 2 mmol/L KCl, 1.8 mmol/L $CaCl_2{\cdot}2H_2O$, 1 mmol/L $MgCl_2{\cdot}6H_2O$, and 5 mmol/L HEPES. Before the uptake experiments, oocytes were preincubated in ND96 containing 1 mmol/L glutarate for 2 h, to generate an intracellular-to-extracellular gradient of glutarate concentration. In all uptake experiments, incubation was performed in ND96 solution containing 18.5 kBq of ^{99m}Tc-MAG3, at 25°C for 60 min, unless otherwise indicated.

In the time course experiments, uptake of ^{99m}Tc-MAG3 was measured with incubation for 10, 30, 60, or 120 min. To assess dependence of ^{99m}Tc-MAG3 transport on extracellular Na^+, uptake of ^{99m}Tc-MAG3 into *Xenopus laevis* oocytes expressing OAT1 was measured in the presence or absence of Na^+ in ND96 (in the absence of NaCl, an equal concentration of choline-Cl was used). To assess dependence of ^{99m}Tc-MAG3 transport on intracellular glutarate, OAT1-expressing *Xenopus laevis* oocytes were preincubated in ND96 with or without 1 mmol/L glutarate for 2 h before being used in uptake experiments. The control oocytes were injected with a volume of water equal to that of the OAT1 cRNA and vehicle. For inhibition experiments, the following inhibitors were used: PAH, OIH, probenecid, 3,5-diiodo-4-pyridone-*N*-acetate (iodopyracet), furosemide, ethacrynic acid, glucoheptonate, L-Tyr, maleic acid, tetraethylammonium (TEA), and 2,4-dinitrophenol (DNP) (*5,12–17*). DNP was used at a concentration of 0.1 mmol/L (*16*); the other inhibitors were used at a concentration of 2 mmol/L. The control oocytes were incubated in the absence of inhibitor.

In all experiments, incubation was terminated by washing with ice-cold ND96 solution 5 times. The radioactivity of each oocyte was measured using a well-scintillation counter (ARC-380; Aloka). Each experiment was repeated 3 times using different batches of *Xenopus laevis* oocytes.

Statistical Analysis

The values obtained in each experiment were expressed as the mean ± SD. Statistical comparisons between groups were performed using the Student unpaired *t* test.

RESULTS

In the preparation of the radiolabeled compound ^{99m}Tc-MAG3, labeling efficiency was greater than 95.0%. After purification, the radiochemical purity was greater than 99.9%. The specific activity of the ^{99m} Tc-MAG3 was 6.6×10^{14} Bq/mol.

As shown in Figure 2, the ^{99m}Tc-MAG3 concentration in OAT1-expressing *Xenopus laevis* oocytes reached a steady state at 60 min after addition of the tracer. Consequently, in the uptake experiments, the oocytes were incubated for 60 min.

Figures 3 and 4 summarize the characterization of ^{99m}Tc-MAG3 transport in OAT1-expressing *Xenopus laevis* oocytes under various incubation conditions. ^{99m}Tc-MAG3 uptake was significantly higher in OAT1-expressing oocytes than in water-injected control oocytes ($P < 0.001$) (Figs. 3 and 4). In general, results obtained in the presence of Na^+ were similar to those obtained in the absence of Na^+ (Fig. 3). In OAT1-expressing oocytes, Na^+ had a slight but significant effect ($P < 0.05$) on ^{99m}Tc-MAG3 uptake (Fig. 3). ^{99m}Tc-MAG3 uptake was higher in oocytes preincubated with glutarate than in oocytes that were not preincubated

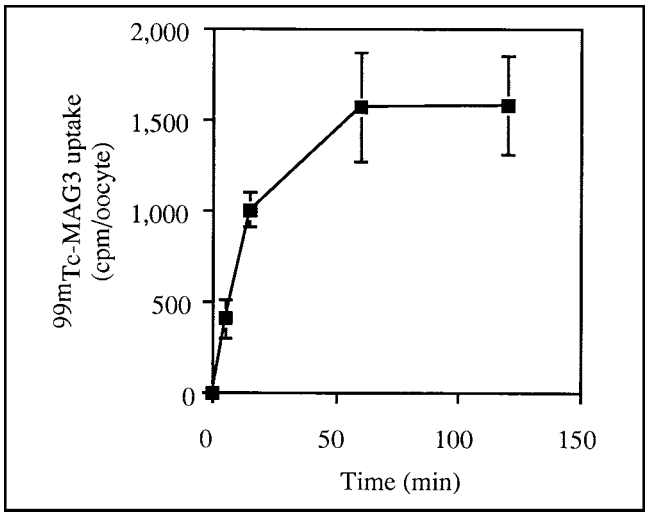

FIGURE 2. Accumulation of ^{99m}Tc-MAG3 in OAT1-expressing *Xenopus laevis* oocytes. Values represent the mean ± SD (n = 6–8 oocytes).

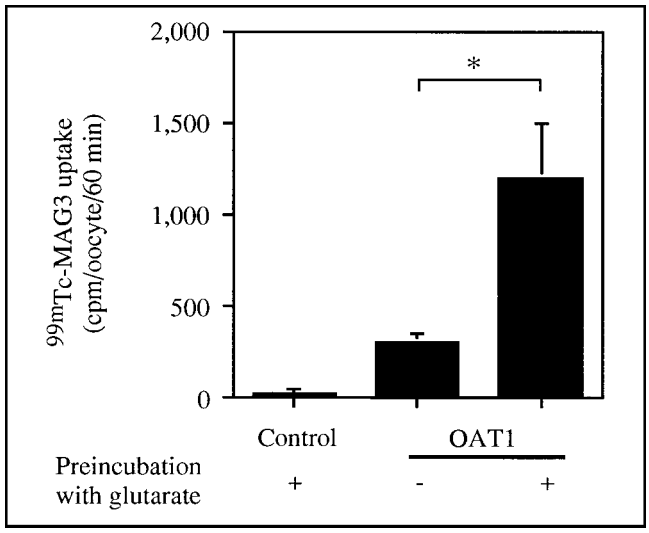

FIGURE 4. ^{99m}Tc-MAG3 transport dependence on intracellular glutarate in OAT1-expressing *Xenopus laevis* oocytes. Control oocytes were injected with same volume of water, instead of OAT1 cRNA. Values represent mean ± SD (n = 15–20 oocytes). $^*P < 0.001$.

with glutarate (Fig. 4). Accumulation of glutarate in *Xenopus laevis* oocytes *trans*-stimulated uptake of ^{99m}Tc-MAG3 via OAT1.

Figure 5 shows the results of the inhibition experiments. ^{99m}Tc-MAG3 uptake was significantly inhibited ($P < 0.001$) by the following compounds: PAH, 8.5% ± 16.2% of ^{99m}Tc-MAG3 uptake in the absence of an inhibitor; OIH, 26.4% ± 21.7%; probenecid, 29.1% ± 12.4%; iodopyracet, 15.8% ± 7.9%; furosemide, 30.5% ± 15.7%; ethacrynic acid, 21.6% ± 10.6%; and glucoheptonate, 35.6% ± 22.6%. TEA, an inhibitor of organic cation transport (*6*), did not inhibit ^{99m}Tc-MAG3 uptake. Other compounds that did not significantly inhibit ^{99m}Tc-MAG3 uptake were DNP (*16*) and L-Tyr. Maleic acid, which causes experimental Fanconi syndrome (*17*), significantly inhibited ^{99m}Tc-MAG3 transport (60.1% ± 18.7%) ($P < 0.01$).

DISCUSSION

Organic anion transport is thought to be an important system for diagnosis of kidney function, because many anions, whether of endogenous or environmental origin, are eliminated from the body by the organic anion secretory system of the renal proximal tubule (*5*), and it has been proposed that this system is involved in drug metabolism

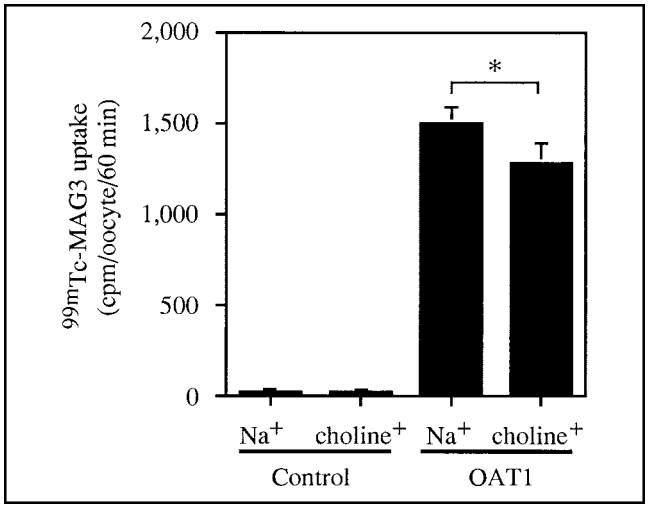

FIGURE 3. ^{99m}Tc-MAG3 transport dependence on extracellular Na^+ in OAT1-expressing *Xenopus laevis* oocytes. Uptake of ^{99m}Tc-MAG3 was measured in the presence of Na^+ (Na^+) or absence of Na^+ ($choline^+$). The control oocytes were injected with same volume of water, instead of OAT1 cRNA. Values represent mean ± SD (n = 15–20 oocytes). $^*P < 0.05$.

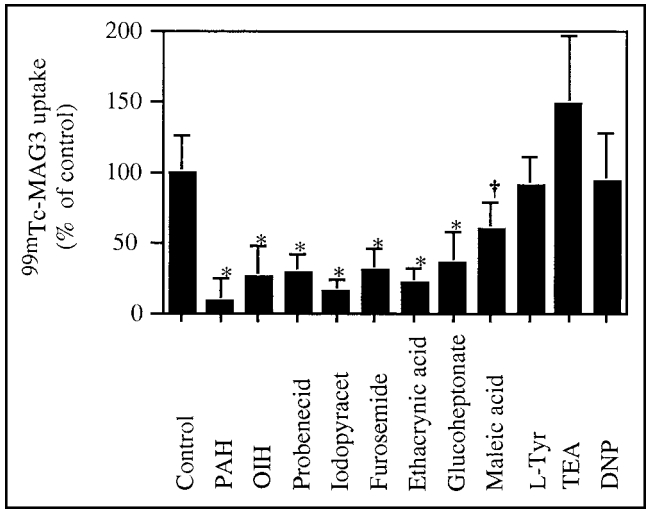

FIGURE 5. Inhibition of OAT-mediated ^{99m}Tc-MAG3 uptake by various drugs. Control value is ^{99m}Tc-MAG3 uptake in absence of inhibitor. ^{99m}Tc-MAG3 uptake in presence of inhibitor is expressed as percentage of control value (mean ± SD; n = 16–22 oocytes). $^*P < 0.001$. $^\dagger P < 0.01$.

and renal toxicity (*6*). Transepithelial excretion of organic anions occurs via accumulative transport of these ions from blood into proximal tubule cells across the basolateral membrane, and subsequent secretion into the urine through the apical membrane (*6*). Based on experiments using isolated segments of proximal tubules, it has been estimated that, in the course of secretion, the intracellular concentration of the organic anion PAH rises to 100–300 times the concentration in either the peritubular or luminal fluid (*18*). Accumulative transport of radiolabeled organic anions such as ^{99m}Tc-MAG3 enables scintigraphic imaging of renal function.

The molecular structure and properties of organic anion transporters have gradually been clarified. In the last decade, there has been great interest in the specificity of the basolateral organic anion transport system (which transports a remarkably broad spectrum of agents) and in the energetics of transport (which is driven by complex tertiary coupling to metabolic energy) (*5*). Recently, OAT1 cDNA was cloned. Figure 1 shows the putative membrane topology of OAT1. OAT1 cDNA consists of 2,294 nucleotides and contains an open reading frame encoding a 551-amino-acid protein with 12 putative membrane-spanning domains (*6*). Human (*19,20*) and mouse (*21,22*) homologues of OAT1 have been cloned and characterized. The characteristics of OAT1 are the same as those of the organic anion/dicarboxylate exchanger predicted by physiologic studies to be located at the basolateral membrane of proximal tubules. Northern blotting and in situ hybridization have revealed that OAT1 is exclusively expressed in particular segments of proximal tubules, presumably the middle segment (*6*). Using an immunohistochemical assay, Tojo et al. detected expression of OAT1 in the basolateral membrane of rat proximal tubule cells (*23*). It is generally assumed that the first step in ^{99m}Tc-MAG3 secretion by proximal tubule cells is extraction of ^{99m}Tc-MAG3 from peritubular plasma by proximal tubule cells through the basolateral membrane. OAT1 transcript has not been detected in rat heart, brain, spleen, lung, liver, skeletal muscle, or testis, but it has been detected (as a strong Northern blot signal) in rat kidney (*7*).

Shimada et al. have proposed that, in the basolateral membrane, 3 transport systems cooperate to accumulate PAH in cells (Fig. 6). The primary active transport system at the basolateral cell membrane is Na^+/K^+-adenosine triphosphatase, which maintains an electrochemical potential difference by transporting Na^+ across the basolateral cell membrane. Intracellular accumulation of certain dicarboxylates far above plasma concentrations occurs as a result of cotransport of dicarboxylates with 3 Na^+ (*24*). In addition, an Na^+-coupled system mediates uptake of Krebs cycle intermediates across the luminal membrane (*25*). This hypothesis is supported by findings indicating that metabolic fuels stimulate accumulation of PAH in renal cortex (*26–28*). Dicarboxylates that accumulate intracellularly (most likely 2-oxoglutarete) are subsequently exchanged with extracellular PAH, providing a possible mechanism for

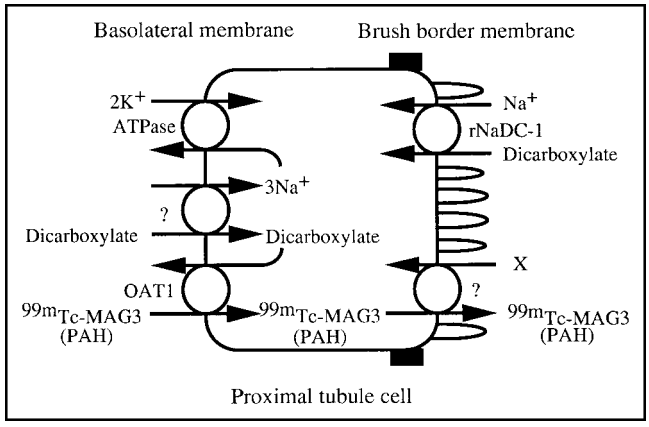

FIGURE 6. Proposed scheme for membrane transport of ^{99m}Tc-MAG3 in proximal tubule cell. According to anion exchange model of renal organic anion transport model, organic anions are transported into cell in exchange for intracellular dicarboxylates, which subsequently return to cell interior via rNaDC-1. In this model, OAT1 is indirect Na^+-coupled organic anion transporter driven by complex tertiary coupling to metabolic energy. ATPase = adenosine triphosphatase.

accumulation of PAH at high levels within cells. PAH may leave the cell via the brush-border membrane, by following its electrochemical gradient. This proposed model is very consistent with the present results.

In the uptake experiment with OAT1-expressing *Xenopus laevis* oocytes, intracellular concentration of ^{99m}Tc-MAG3 was 200.8 ± 75.6 times its extracellular concentration (n = 22). This indicates that uptake of ^{99m}Tc-MAG3 is due to active transport (tertiary active transport in Fig. 6) against an outwardly directed ^{99m}Tc-MAG3 concentration gradient, rather than facilitated diffusion. In a study using isolated segments, a similar level of accumulation of PAH was observed (*18*).

Figures 2 and 3 show that ^{99m}Tc-MAG3 acts as a substrate of OAT1 in OAT1-expressing *Xenopus laevis* oocytes. ^{99m}Tc-MAG3 transport via OAT1 is slightly but significantly dependent on extracellular Na^+ concentration ($P < 0.05$) (Fig. 3). This suggests that Na^+ indirectly contributes to ^{99m}Tc-MAG3 transport.

Figure 4 shows the effect of preincubation with glutarate on transport of ^{99m}Tc-MAG3 by OAT1-expressing *Xenopus laevis* oocytes. Accumulated glutarate stimulates ^{99m}Tc-MAG3 uptake via OAT1. The *trans*-stimulative effect of glutarate indicates that ^{99m}Tc-MAG3 acts as a substrate of OAT1, which is an organic anion/dicarboxylate exchanger. It has been reported that coexpression of OAT1 and rat Na^+ dicarboxylate transporter (rNaDC-1) in *Xenopus laevis* oocytes results in greater PAH uptake than does expression of OAT1 only, because of the outwardly directed dicarboxylate gradient created by rNaDC-1 (*6*). The anion exchange model suggests that, at the basolateral membrane, ^{99m}Tc-MAG3 is also exchanged with anions generated during cellular metabolic processes including the Krebs cycle; for example, lactate or dicarboxylates.

On the other hand, it is important to know which drugs affect ^{99m}Tc-MAG3 accumulation in kidneys. We tested several drugs in the present study. In human studies, PAH loading with infusion caused a significant decrease in ^{99m}Tc-MAG3 clearance (*3*). Figure 5 shows that both PAH and OIH (induced hippurates) inhibited ^{99m}Tc-MAG3 OAT1-mediated transport ($P < 0.001$). Probenecid, which is a renal secretion blocker and is also used as a uricosuric drug (*13*), inhibited renal clearance of ^{99m}Tc-MAG3 ($P < 0.001$) (*29*). OAT1-mediated ^{99m}Tc-MAG3 uptake was also inhibited ($P < 0.001$) by iodopyracet, which has long been used as a radiographic contrast medium. In general, loop diuretics (such as ethacrynic acid and furosemide) are bound to plasma protein (*18*). Also, results of micropuncture studies indicate that most diuretics are not filtered at the glomerulus (*18*). Thus, a possible mechanism of appearance of diuretics in proximal tubules is secretion into proximal tubules via active transport (*18*). Furosemide and ethacrynic acid had *cis*-inhibitory effects on ^{99m}Tc-MAG3 uptake in the present study ($P < 0.001$). These compounds had a similar effect on ^{14}C-PAH uptake in a study by Sekine et al. (*6*). It appears that secretion of potent loop diuretics into the urine of the pars recta may be an important feature of their mode of action (*18*). Figure 5 shows that both glucoheptonate and ^{99m}Tc-MAG3 are transported by OAT1. Glucoheptonate, an organic anion, is a ligand of ^{99m}Tc-glucoheptonate (*30*). Maleic acid, which causes experimental Fanconi syndrome (*17*), had a significant inhibitory effect ($P < 0.01$).

TEA, which has previously been found to inhibit organic cation transport, did not inhibit ^{99m}Tc-MAG3 transport (*5*). L-Tyr (a neutral amino acid) and ^{99m}Tc-MAG3 are not both transported by OAT1. Unexpectedly, ^{99m}Tc-MAG3 transport was not significantly inhibited by DNP, which inhibits generation of high-energy phosphates (*16*); organic anion transport is thought to involve a tertiary active transport system, as described above. Other important clinically relevant drugs that inhibit transport via OAT1 include penicillin and thiazide diuretics. Further investigation of the effects of these compounds on ^{99m}Tc-MAG3 transport is needed.

The most conspicuous feature of this transport system is its extremely wide substrate selectivity, which includes not only endogenous anions but also several clinically important drugs (*6*). For example, it has been reported that OAT1 transports the following compounds: diclofenac, diflunisal, flurbiprofen, ibuprofen, indomethacin, ketoprofen, meclofenamate, naproxen, oxyphenbutazone, phenacetin, phenylbutazone, piroxicam, tolmetin (hydrophobic nonsteroidal antiinflammatory drugs [NSAIDs]), and salicylate (hydrophilic NSAIDs) (*31*). Transport of ^{14}C-PAH via OAT1 is strongly inhibited by cephaloridine (β-lactam antibiotic), nalidixic acid (old quinolone), valproic acid (antiepileptic drug), α-ketoglutarate, and urate (*6*). There have been reports of the substrate structure of organic anion transporters. Binding of organic anions with the renal organic anion transporter depends mainly on hydrophobic interactions, hydrogen bonding, and electrostatic interactions between the substrate and the carrier (*32*). The organic anion transporter interacts with substrates that contain a hydrophobic core with negative charges or negative partial charges (*33*). The organic anion transport system requires only a hydrophobic backbone and negative or partial negative charges optimally separated by 60–70 nm (*34*). The substrates of OAT1 are apparently structurally unrelated to each other. These previous findings provide a possible explanation for the extremely wide substrate selectivity of OAT1.

To date, 4 isoforms of OAT have been identified: OAT1, OAT2 (*8*), OAT3 (*9,10*), and OAT4 (*35*). Transcellular transport of organic anions has been demonstrated in various tissues, including kidney, brain, liver, and placenta. Incidentally, sporadic liver and gallbladder visualization has been observed during studies of renal transport of ^{99m}Tc-MAG3, and fasting increases hepatobiliary excretion (*11*). Because ^{99m}Tc-MAG3 is dianionic at physiologic pH, it is likely that it is transported by hepatocytes using the anionic transport system at physiologic pH (*11*). OAT2 is expressed strongly in the liver and weakly in the kidney; OAT3 is expressed strongly in the liver, kidney, and brain and weakly in the eye. Indocyanine green has been found to inhibit transport of substrates such as PAH via both OAT2 and OAT3 (*8,9*). Furthermore, blocking the anionic transport system of the liver by infusing rats with indocyanine green before injecting ^{99m}Tc-MAG3 has been found to significantly decrease the amount of ^{99m}Tc-MAG3 excreted into the bile (*11*). This suggests a connection between ^{99m}Tc-MAG3 excretion and PAH excretion via the isoforms of OAT in the hepatobiliary system.

Numerous organic anions, including ^{99m}Tc-MAG3, are efficiently excreted from renal proximal tubules via carrier-mediated pathways. In the basolateral membrane, the present results indicate that ^{99m}Tc-MAG3 is a substrate of OAT1, whereas ^{99m}Tc-MAG3 transport in the brush border membrane is less clearly understood (Fig. 6). We believe that transport experiments using *Xenopus laevis* oocytes can help to lay the foundation for development of improved pharmaceuticals for evaluation of kidney function. We anticipate future studies of ^{99m}Tc-MAG3 transport via other isoforms of OAT, including analysis of relative kinetics.

CONCLUSION

PAH, OIH, probenecid, iodopyracet, furosemide, ethacrynic acid, and glucoheptonate had *cis*-inhibitory effects on ^{99m}Tc-MAG3 transport. Glutarate had a *trans*-stimulative effect. The present results are consistent with the proposed model of organic anion transport into cells in exchange for intracellular dicarboxylates, which subsequently return to the cell interior via a rNaDC-1. Thus, ^{99m}Tc-MAG3 acts as a substrate of OAT1, an organic anion/dicarboxylate exchanger.

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