MATE1 and MATE2K Transporter

Introduction: The human multidrug and toxin extrusion family transporters MATE1 (SLC47A1) and MATE2K (SLC47A2) are well characterized transporters acting as H+/organic cation antiporters. Human MATE1 is highly expressed in the liver at the canalicular membrane of hepatocytes and in the kidney at the apical side of renal proximal and distal tubule cells whereas MATE2K is kidney-specific localized at the brush-border membrane of proximal tubules. MATE1 and MATE2K are involved in the excretion of important medications and the disruption of these transporters can cause severe pharmacological problems. Therefore, it is important to evaluate the interaction of new chemical entities with these transporters. Typical substrates for MATEs are hydrophilic, low-molecular-weight organic cations such as metformin and 1-methy-4-phenylpyridinium (MPP), but they also transport a wide range of other compounds like acyclovir, cimetidine, oxaliplatin, and fexofenadine. For MATE2K the regulatory agency EMA require *in vitro* evaluation for drug candidates that are eliminated via the kidneys. For liver eliminated drugs in vitro studies with MATE1 are recommended by EMA.

Methods: PortaCellTec generated HEK293 cell lines stably transfected with MATE1 or MATE2-K transporter proteins and validated the cell-transporter system with two probe substrates (^{14}C-metformin and ^{3}H-1-methyl-4-phenylpyridinium (MPP)) and three inhibitors (Cimetidine, Ketoconazole and Quinidine). To perform uptake experiments, transporter-transfected and control cells were harvested, plated into 24-well-plates and were cultured for 3 days. To generate an intracellular acidification the cells were preincubated for 30 min in a 30 mM NH_4Cl solution at pH 7.4 and 37°C. The uptake was initiated by adding the probe substrate in the absence and presence of an inhibitor. After 1 min the uptake was terminated by washing three times with cold assay buffer. The radio-labeled content of each cell lysates was analyzed by liquid scintillation counting.

MATE1 - SLC47A1

Substrate	Inhibitor	Kinetic parameters	References
MPP	---	K_m = 89 µM	K_m = 100 µM (Tanihara, 2007) K_m = 12 µM (Dangprapai, 2011)
Metformin	---	K_m = 274 µM	K_m = 227 µM (Chen, 2009) K_m = 202 µM (Meyer, 2010)
MPP	Cimetidine	IC_{50} = 9.7 µM	K_i = 2.7 µM (Ito, 2012)
Metformin	Cimetidine	IC_{50} = 1.8 µM	K_i = 3.8 µM (Ito, 2012)

MATE2K - SLC47A2

Substrate	Inhibitor	Kinetic parameters	References
MPP	---	K_m = 68 µM	K_m = 110 µM (Tanihara, 2007) K_m = 94 µM (Masuda, 2006)
Metformin	---	K_m = 934 µM	K_m = 1980 µM (Tanihara, 2007) K_m = 1050 µM (Masuda, 2006)
MPP	Cimetidine	IC_{50} = 24 µM	K_i = 4.0 µM (Ito, 2012)
Metformin	Cimetidine	IC_{50} = 5.7 µM	K_i = 6.9 µM (Ito, 2012)

Figure 1 Concentration dependent MATE1 mediated net-uptake of MPP and metformin

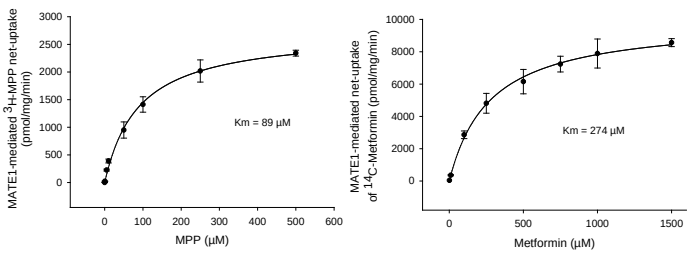


Figure 2 Concentration dependent MATE2K mediated net-uptake of MPP and metformin

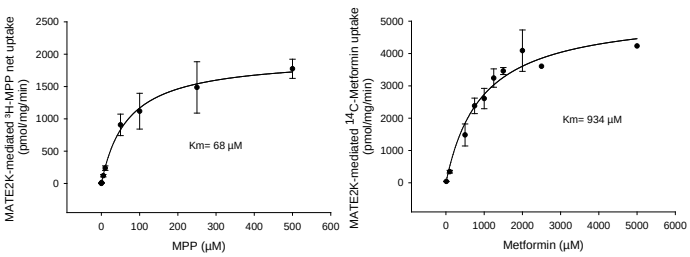


Figure 3 Inhibition of MATE1 mediated MPP net-uptake by cimetidine

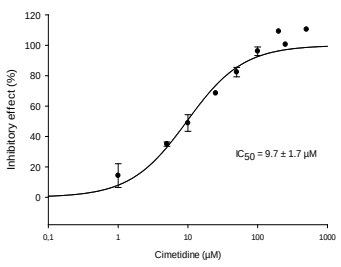


Figure 4 Inhibition of MATE1 mediated metformin net-uptake by cimetidine

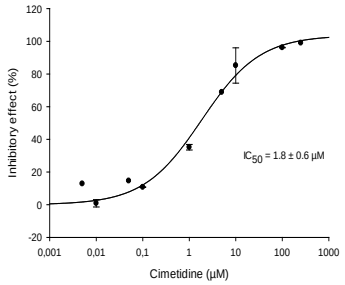


Figure 5 Inhibition of MATE2K mediated MPP net-uptake by cimetidine

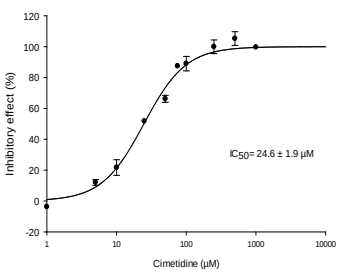
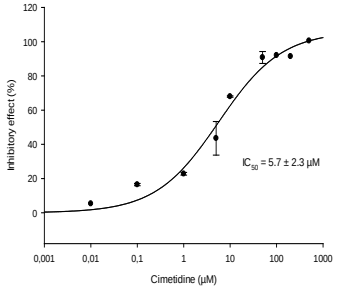


Figure 6 Inhibition of MATE2K mediated metformin net-uptake by cimetidine



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