PEPT2

The human peptide transporter 2, PEPT2, is a low-capacity/high-affinity proton-coupled cotransporter of diverse di- and tri-peptides as well as peptidomimetic substrates, and is expressed in a variety of tissues, particularly in the kidney and brain. PEPT2 is primarily involved in the renal reabsorption of di-/tripeptides and drugs with peptide-like structures. In the kidney, PEPT2 function is complemented by PEPT1, a closely related, high-capacity/low-affinity transporter with predominant expression in small intestine. The efficient reabsorption of peptide-bound amino nitrogen at the apical membrane of renal proximal tubular epithelium is an important process in systemic nitrogen homeostasis.

Among the genes involved in the porphyrin synthesis pathway, the mRNA expression of Peptide transporter 2 (PEPT2) in FGS fluorescence-positive gliomas was significantly higher than that in fluorescence-negative gliomas. Protein expression of PEPT2 was also significantly higher in the fluorescence-positive gliomas, which was confirmed by western blot analysis and immunofluorescence analysis. The siRNA-mediated downregulation of the mRNA and protein expression of PEPT2 led to decreased PpIX fluorescence intensity, as confirmed by fluorescence spectrum analysis.

The results suggest PEPT2 is an important candidate molecule in 5-ALA-mediated FGS in grade II/III gliomas. As the overexpression of PEPT2 was associated with higher PpIX fluorescence intensity, PEPT2 may improve fluorescence-guided resection in grade II/III gliomas [1)].

The aims of the current study were (1) to quantify the role of PEPT2 in the uptake of glycylsarcosine (GlySar) in cultured neonatal astrocytes and (2) to examine GlySar transport and PEPT2 expression in two glioma cell lines. The uptake of [(14)C]GlySar was measured in astrocytes cultured from neonatal mouse (PEPT2(+/+) and PEPT2(-/-)) and rat, as well as rat C6 and F98 glioma cells. PEPT2 expression was examined by reverse transcription-polymerase chain reaction (RT-PCR). Neonatal astrocytes from PEPT2(-/-) mice had a 94% reduction in [(14)C]GlySar uptake compared to wild type mice and there was no saturable transport. In PEPT2(+/+) mice, [(14)C]GlySar uptake was saturable (V(max) 58 +/- 12 pmol/mg/min, K(m) 107 +/- 46 microM, K(d) 0.043 +/- 0.004 microl/mg/min). In neonatal rat astrocytes, kinetic analysis also suggested that [(14)C]GlySar uptake was via a single transporter. The inhibitor profile and pH dependence of that transport process was consistent with PEPT2. In C6 and F98 glioma cells, [(14)C]GlySar uptake was markedly reduced (approximately 96-98%) compared to that in neonatal astrocytes and this was reflected by an absence of PEPT2 mRNA expression. These results indicate that PEPT2 is the sole transporter involved in the uptake of GlySar into neonatal cultured astrocytes. However, PEPT2 mRNA appears to be absent from two glioma cell lines [2)].

5-Aminolevulinic acid (5-ALA) is a precursor of porphyrins and heme that has been implicated in the neuropsychiatric symptoms associated with porphyrias. It is also being used clinically to delineate malignant gliomas. The blood-CSF barrier may be an important interface for 5-ALA transport between blood and brain as in vivo studies have indicated 5-ALA is taken up by the choroid plexuses whereas the normal blood-brain barrier appears to be relatively impermeable. This study examines the mechanisms of 5-[(3)H]ALA uptake into isolated rat lateral ventricle choroid plexuses. Results suggest

that there are two uptake mechanisms. The first was a Na(+)-independent uptake system that was pH dependent (being stimulated at low pH). Uptake was inhibited by the dipeptide Gly-Gly and by cefadroxil, an alpha-amino-containing cephalosporin. These properties are the same as the proton-dependent peptide transporters PEPT1 and PEPT2, which have recently been shown to transport 5-ALA in frog oocyte expression experiments. Choroid plexus uptake was not inhibited by captopril, a PEPT1 inhibitor, suggesting PEPT2-mediated uptake. The presence of PEPT2 and absence of PEPT1 in the choroid plexus were confirmed by western blotting. The second potential mechanism was both Na(+) and HCO(3)(-) dependent and appears to be an organic anion transporter, although it is possible that removal of Na(+) and HCO(3)(-) may indirectly affect PEPT2 by affecting intracellular pH. The presence of PEPT2 and a putative Na(+)/HCO(3)(-)-dependent organic anion transporter is important not only for an understanding of 5-ALA movement between blood and brain but also because these transporters may affect the distribution of a number of drugs between blood and CSF [3].

Unclassified

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