

Polyclonal Antibody to Peptide Transporter 1 (PepT1) - Aff - Purified

Catalog No.:	R1532P
Quantity:	0.1 mg
Concentration:	1.0 mg/ml (by UV absorbance at 280 nm)
Background:	<p>Peptide Transporter 1 (PepT1) is an oligopeptide transporter expressed at the brush-border membrane of intestinal epithelial cells and has a predominant role in intestinal absorption of natural di- and tripeptides and peptide-based drugs. PepT1 is localized predominantly in the upper small intestine, with decreasing expression in the ileum. PepT1 is a H(+)-coupled transporter that is able to transport oligopeptides and peptide-derived antibiotics through the small intestine. PepT1-mediated uptake is independent of extracellular Na⁺, K⁺ and Cl⁻, and of membrane potential. PepT1 displays low affinity and unusually broad substrate specificity. Consequently, PepT1 accepts various peptide mimetic drugs such as β-lactam antibiotics, angiotensin converting enzyme inhibitors, renin inhibitors and anticancer drugs. PepT1 can be exploited for improvement of intestinal absorption of poorly absorbed drugs by derivation of the drugs to peptide mimetics. Such strategies have been tested across the membranes of human intestinal epithelial-like Caco-2 cells and in certain tumor cells. PepT1 may improve delivery of peptide-mimetic anticancer drugs to tumors by utilization of this peptide transporter activity. Lys-Xaa dipeptides and derivatives have been shown to be an effective competitive inhibitor of mammalian PepT1 in Caco-2 cells.</p>
Host:	Rabbit
Immunogen:	Synthetic peptide C-T-M-S-G-K-I-G-A-L-E-I corresponding to human PepT1 conjugated to KLH using maleimide. A residue of cysteine was added to the carboxy terminal end to facilitate coupling.
Format:	<p>State: Liquid (sterile filtered) purified Ig fraction. Purification: Affinity purified from whole rabbit serum Buffer System: 0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2, containing 0.01% (w/v) Sodium Azide as preservative</p>
Applications:	<p>Affinity purified rabbit anti-PepT1 has been tested by ELISA, immunohistochemistry and western blotting against both the immunizing peptide, native and recombinant proteins. Although not tested, this antibody is likely functional for immunoprecipitation and other immunodetection techniques.</p> <p>This product was assayed by immunoblot and was found to be reactive at 1:2,000 with extracts from human liver and small intestine.</p> <p>Strong immunoreactivity was observed in similar tissue when stained for immunohistochemistry using a 1:2,000 dilution of the antibody.</p> <p>Other applications not tested. Optimal dilutions are dependent on conditions and should be determined by the user.</p>

- Specificity:** This antibody is directed against human PepT1 and is useful in determining its presence in various assays. Anti-PepT1 antibody detects endogenous and overexpressed proteins found in various tissues and extracts. Reactivity is observed against human PepT1 and cross reactivity with PepT1 from other mammalian sources has not been tested.
- Storage:** Store (in aliquots) at -20°C or below. Avoid repeated freezing and thawing. Dilute only prior to immediate use. Shelf life: one year from despatch.
- General Readings:** C.J. Sword, A.A. Constantinou, I.D. Collier, P.D. Bailey and C.A.R. Boyd (2000) Modified dipeptides inhibit substrate uptake into PepT1-expressing *Xenopus* oocytes. *J. Physiology*: 528P, 99.
Fei, Y.J., Kanai, Y., Nussberger, S., Ganapathy, V., Leibach, F.H., Romero, M.F., Singh, S.K., Boron, W.F. & Hediger, M.A. (1994). Expression cloning of a mammalian proton-coupled oligopeptide transporter. *Nature* 368, 563-566.
D.R. Herrera, Q. Wang, O.S. Gudmundsson, T. Cook, R. Smith, T.N. Faria, and G.T. Knipp. (2001) Spatial Expression Patterns of Peptide Transporters in the Human and Rat Gastrointestinal Tracts, Caco-2 in vitro Cell Culture, and Multiple Human Tissues. *AAPS PharmSci.* 3(1):9.
D.R. Herrera and G.T. Knipp (2003). Current Perspectives on Established and Putative Mammalian Oligopeptide Transporters. *J. Pharm. Sci.* 92:000-000.

- Pictures:** **Figure 1.** Anti-PepT1 polyclonal antibody detects human PepT1 by Western blot. Lane 1 - human small intestine lysate, Lane 2 - human kidney lysate, Lane 3 - human duodenum lysate, Lane 4 - human liver lysate. The apparent molecular weight of PepT1 in human liver is 90 kDa. The expression in liver, but not other tissues is consistent with RT-PCR experiments described in Herrera et al. A 10% gel was used to separate the protein by SDS-PAGE. The protein was transferred to nitrocellulose using standard methods. After blocking the membrane was probed with a 1:1,000 dilution of the primary antibody for 1 h at room temperature followed by washes and reaction with a 1:4000 dilution of HRP conjugated Dnky-a-Rabbit IgG [H&L] for 30 min at room temperature. Signal detection occurred using Pierce Chemical's Femto signal kit. Other detection systems will yield similar results.

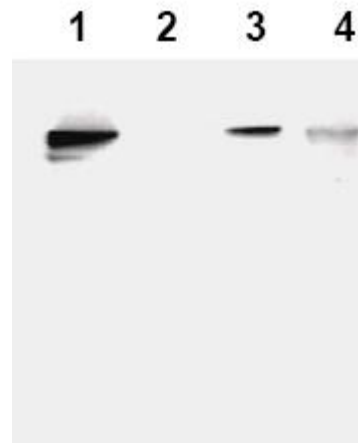


Figure 3. Immunohistochemical staining after sectioning and deparaffination of human liver tissue using (A) 1:2,000 dilution of anti-PepT1 antibody and (B) pre-immune (control) sera followed by processing with HRP Donkey anti-Rabbit IgG [H&L] and chromogenic substrate.

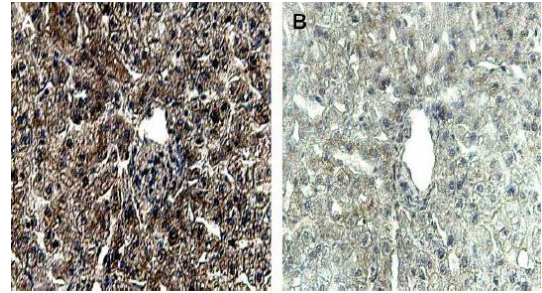


Figure 2. Immunohistochemical staining after sectioning and deparaffination of human small intestine tissue using (A) 1:2,000 dilution of anti-PepT1 antibody and (B) pre-immune (control) sera followed by processing with HRP Donkey anti-Rabbit IgG [H&L] and chromogenic substrate.

