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PEPT1 siRNA (m): sc-156081

BACKGROUND

In mammalian small intestine, the proton-coupled peptide transporter (PEPT) is responsible for the absorption of small peptides arising from digestion of dietary proteins. PEPT1, a hydrogen ion/peptide cotransporter, transports dipeptides and tripeptides, but not free amino acids or peptides with more than three amino acid residues. Its driving force for uphill transport requires proton binding and the presence of an inside-negative membrane potential. PEPT1 is 708 amino acid protein that contains 12 putative membrane-spanning domains. PEPT1 is expressed in Caco-2 cells. PEPT1 seems to play important roles in nutritional and pharmacological therapies. The mammalian kidney expresses a proton-coupled peptide transporter, PEPT2, that is responsible for the absorption of small peptides, as well as β -lactam antibiotics and other peptide-like drugs, from the tubular filtrate. The gene which encodes PEPT1 maps to human chromosome 13q32.3.

REFERENCES

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- Liu, W., Liang, R., Ramamoorthy, S., Fei, Y.J., Ganapathy, M.E., Hediger, M.A., Ganapathy, V. and Leibach, F.H. 1995. Molecular cloning of PEPT2, a new member of the H⁺/peptide cotransporter family, from human kidney. *Biochim. Biophys. Acta* 1235: 461-466.
- Adibi, S.A. 1997. The oligopeptide transporter (PEPT1) in human intestine: biology and function. *Gastroenterology* 113: 332-340.
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CHROMOSOMAL LOCATION

Genetic locus: Slc15a1 (mouse) mapping to 14 E5.

PRODUCT

PEPT1 siRNA (m) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PEPT1 shRNA Plasmid (m): sc-156081-SH and PEPT1 shRNA (m) Lentiviral Particles: sc-156081-V as alternate gene silencing products.

For independent verification of PEPT1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-156081A, sc-156081B and sc-156081C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PEPT1 siRNA (m) is recommended for the inhibition of PEPT1 expression in mouse cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

PEPT1 (E-3): sc-373742 is recommended as a control antibody for monitoring of PEPT1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PEPT1 gene expression knockdown using RT-PCR Primer: PEPT1 (m)-PR: sc-156081-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.