



PROT siRNA (m): sc-152483

BACKGROUND

The GAT1 gene family includes sodium- and chloride-dependent plasma membrane transporters for neurotransmitters, metabolites and osmolites, which couple substrate flux to transmembrane electrochemical gradients. PROT (sodium-dependent proline transporter), also known as Solute carrier family 6 member 7, is a 636 amino acid multi-pass membrane protein that is a GAT1 family member specifically expressed in regions of the brain. PROT terminates the action of proline by its high affinity sodium/chloride-dependent reuptake into pre-synaptic terminals. Enriched in glutamatergic synaptic terminals, it is likely that PROT plays an important role in excitatory events of neurotransmission. PROT-mediated proline uptake is inhibited by compounds such as bntropine, LP-403812 and Des-Tyr-Leu-enkephalin (GGFL). These inhibitors of proline uptake may lead to the development of therapeutic agents for certain neurologic disorders.

REFERENCES

- Shafqat, S., et al. 1995. Human brain-specific L-proline transporter: molecular cloning, functional expression, and chromosomal localization of the gene in human and mouse genomes. *Mol. Pharmacol.* 48: 219-229.
- Fremeau, R.T., et al. 1996. A novel nonopioid action of enkephalins: competitive inhibition of the mammalian brain high affinity L-proline transporter. *Mol. Pharmacol.* 49: 1033-1041.
- Galli, A., et al. 1999. L-proline and L-pipecolate induce enkephalin-sensitive currents in human embryonic kidney 293 cells transfected with the high-affinity mammalian brain L-proline transporter. *J. Neurosci.* 19: 6290-6297.
- Jayanathi, L.D., et al. 2000. Differential regulation of mammalian brain-specific proline transporter by calcium and calcium-dependent protein kinases. *Br. J. Pharmacol.* 129: 465-470.
- Igarashi, Y., et al. 2000. Molecular cloning and characterization of a cDNA encoding proline transporter in rice. *Plant Cell Physiol.* 41: 750-756.
- Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 606205. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
- Bröer, S. 2006. The SLC6 orphans are forming a family of amino acid transporters. *Neurochem. Int.* 48: 559-567.
- Anas, M.K., et al. 2008. SIT1 is a betaine/proline transporter that is activated in mouse eggs after fertilization and functions until the 2-cell stage. *Development* 135: 4123-4130.
- Yu, X.C., et al. 2009. Discovery and characterization of potent small molecule inhibitors of the high affinity proline transporter. *Neurosci. Lett.* 451: 212-216.

CHROMOSOMAL LOCATION

Genetic locus: Slc6a7 (mouse) mapping to 18 E1.

PROTOCOLS

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

PRODUCT

PROT 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 PROT shRNA Plasmid (m): sc-152483-SH and PROT shRNA (m) Lentiviral Particles: sc-152483-V as alternate gene silencing products.

For independent verification of PROT (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-152483A, sc-152483B and sc-152483C.

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

PROT siRNA (m) is recommended for the inhibition of PROT 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PROT gene expression knockdown using RT-PCR Primer: PROT (m)-PR: sc-152483-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.