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NTE shRNA (m) Lentiviral Particles: sc-44514-V

BACKGROUND

Neuropathy target esterase (NTE) is a member of a newly discovered protein family, with a domain conserved through evolution. It is an integral membrane protein present in all neurons and in some non-neural cell types of vertebrates. NTE is important in neural development and has the capacity to hydrolyse esters. It is important in the cell-signalling pathway controlling interactions between neurons and accessory glial cells in nervous system development. NTE can be modified by organophosphates, which can cause neuropathy (characterized by axonal degeneration) in humans. NTE loss can lead to prominent neuronal pathology in the thalamus and hippocampus and can also lead to defects in the cerebellum.

REFERENCES

1. Tormo, N., et al. 1993. Soluble and particulate organophosphorus neuropathy target esterase in brain and sciatic nerve of the hen, cat, rat, and chick. *J. Neurochem.* 61: 2164-2168.
2. Quistad, G.B., et al. 2003. Evidence that mouse brain neuropathy target esterase is a lysophospholipase. *Proc. Natl. Acad. Sci. USA* 100: 7983-7987.
3. Li, Y., et al. 2003. Protein domains, catalytic activity, and subcellular distribution of neuropathy target esterase in Mammalian cells. *J. Biol. Chem.* 278: 8820-8825.
4. Akassoglou, K., et al. 2004. Brain-specific deletion of neuropathy target esterase/swisscheese results in neurodegeneration. *Proc. Natl. Acad. Sci. USA* 101: 5075-5080.
5. Zaccheo, O., et al. 2004. Neuropathy target esterase and its yeast homologue degrade phosphatidylcholine to glycerophosphocholine in living cells. *J. Biol. Chem.* 279: 24024-24033.

CHROMOSOMAL LOCATION

Genetic locus: Pnpla6 (mouse) mapping to 8 A1.1.

PRODUCT

NTE shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see NTE siRNA (m): sc-44514 and NTE shRNA Plasmid (m): sc-44514-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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

APPLICATIONS

NTE shRNA (m) Lentiviral Particles is recommended for the inhibition of NTE expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

NTE (K-19): sc-30587 is recommended as a control antibody for monitoring of NTE 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NTE gene expression knockdown using RT-PCR Primer: NTE (m)-PR: sc-44514-PR (20 μ l). Annealing temperature for the primers should be $55-60^{\circ}$ C and the extension temperature should be $68-72^{\circ}$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.