SANTA CRUZ BIOTECHNOLOGY, INC.

NTN siRNA (h): sc-41968

BACKGROUND

Glial cell line-derived neurotrophic factor (GDNF) and the related neurotrophic factor neurturin (NTN) are potent survival factors for central and peripheral neurons. Two receptors for these factors, GDNFRα (also designated RETL1 or TrnR1) and RETL2 (also designated TrnR2), have been identified. The receptors do not contain transmembrane domains and are attached to the cell membrance by glycosyl-phosphoinositol linkage. Binding of GDNF or NTN to either of these receptors leads to the activation of the tyrosine kinase Ret. GDNF and NTN have also been shown to be capable of activating the MAP kinase and PI 3-kinase pathways, indicating a potential role for these proteins in neuronal survival and in the development of many neuronal populations.

REFERENCES

1. Lin, L.F., et al. 1993. GDNF: a glial cell line-derived neurotrophic factor for midbrain dopaminergic neurons. Science 260: 1130-1132.
2. Kotzbauer, P.T., et al. 1996. Neurturin, a relative of glial-cell-line derived neurotrophic factor. Nature 384: 467-470.
3. Jing, S., et al. 1997. GFRα-2 and GFRα-3 are two new receptors for ligands of the GDNF family. J. Biol. Chem. 272: 33111-33117.
4. Widenfalk, J., et al. 1997. Neurturin and glial cell line-derived neurotrophic factor receptor-β (GDNFRβ), novel proteins related to GDNF and GDNFRα with specific cellular patterns of expression suggesting roles in the developing and adult nervous system and in peripheral organs. J. Neurosci. 17: 8506-8519.
5. Sanicola, M., et al. 1997. Glial cell line-derived neurotrophic factor-dependent RET activation can be mediated by two different cell-surface accessory proteins. Proc. Natl. Acad. Sci. USA 94: 6238-6243.
6. Creedon, D.J., et al. 1997. Neurturin shares receptors and signal transduction pathways with glial cell line-derived neurotrophic factor in sympathetic neurons. Proc. Natl. Acad. Sci. USA 94: 7018-7023.
7. Baloh, R.H., et al. 1997. TrnR2, a novel receptor that mediates neurturin and GDNF signaling through Ret. Neuron 18: 793-802.

CHROMOSOMAL LOCATION

Genetic locus: NRTN (human) mapping to 19p13.3.

PRODUCT

NTN siRNA (h) is a pool of 2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NTN shRNA Plasmid (h): sc-41968-SH and NTN shRNA (h) Lentiviral Particles: sc-41968-V as alternate gene silencing products.

For independent verification of NTN (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-41968A and sc-41968B.

RESEARCH USE

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

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

NTN siRNA (h) is recommended for the inhibition of NTN expression in human 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 µM in 66 µl. 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

NTN (D-10): sc-393626 is recommended as a control antibody for monitoring of NTN 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 NTN gene expression knockdown using RT-PCR Primer: NTN (h)-PR: sc-41968-PR (20 µl, 491 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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