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Neurotensin siRNA (h): sc-42115

BACKGROUND

The Neurotensin precursor (also known as Pro-NT/NMN) is a 170 amino acid protein that is cleaved at carboxy terminal dibasic residues by prohormone convertase to generate three processed peptides, designated Neurotensin (NT), Neuromedin N (also known as NMN) and large Neuromedin N (also known as NMN-125). The Neurotensin precursor is processed in the central nervous system and in the intestine to generate the 13 amino acid processed Neurotensin peptide. While the processed Neurotensin peptide functions as a neurotransmitter to modulate dopaminergic signaling pathways in the brain, it acts as a parahormone that may contribute to the growth of human colon cancers in the gut. The processed peptides Neurotensin and Neuromedin N bind to the G protein-coupled Neurotensin receptor (NTR), resulting in the hydrolysis of phosphatidylinositols and the mobilization of calcium.

REFERENCES

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2. Shi, W.X., et al. 1992. Actions of Neurotensin: a review of the electrophysiological studies. *Ann. N.Y. Acad. Sci.* 668: 129-145.
3. de Nadai, F., et al. 1993. Biosynthesis and posttranslational processing of the Neurotensin/Neuromedin N precursor in the rat medullary thyroid carcinoma 6-23 cell line. Effect of dexamethasone. *Endocrinology* 132: 1614-1620.
4. de Nadai, F., et al. 1994. Post-translational processing of the Neurotensin/Neuromedin N precursor in the central nervous system of the rat—I. Biochemical characterization of maturation products. *Neuroscience* 60: 159-166.
5. Vincent, J.P. 1995. Neurotensin receptors: binding properties, transduction pathways, and structure. *Cell. Mol. Neurobiol.* 15: 501-512.
6. Barbero, P., et al. 1998. PC5-A-mediated processing of pro-Neurotensin in early compartments of the regulated secretory pathway of PC5-transfected PC12 cells. *J. Biol. Chem.* 273: 25339-25346.

CHROMOSOMAL LOCATION

Genetic locus: NTS (human) mapping to 12q21.31.

PRODUCT

Neurotensin siRNA (h) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfactions. Also see Neurotensin shRNA Plasmid (h): sc-42115-SH and Neurotensin shRNA (h) Lentiviral Particles: sc-42115-V as alternate gene silencing products.

For independent verification of Neurotensin (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-42115A, sc-42115B and sc-42115C.

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

Neurotensin siRNA (h) is recommended for the inhibition of Neurotensin 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

Neurotensin (E-5): sc-377503 is recommended as a control antibody for monitoring of Neurotensin 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_x BP-HRP: sc-516102 or m-IgG_x 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_x BP-FITC: sc-516140 or m-IgG_x 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 Neurotensin gene expression knockdown using RT-PCR Primer: Neurotensin (h)-PR: sc-42115-PR (20 µl). 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.