SANTA CRUZ BIOTECHNOLOGY, INC.

NCS-1 siRNA (h): sc-36019

BACKGROUND

NCS-1 (for neuronal calcium sensor-1, also designated frequenin) belongs to a highly conserved family of EF-hand-containing Ca^{2+}-binding proteins expressed mainly in neurons. NCS-1 is localized to neuronal cell bodies and axons throughout the brain and spinal cord. It is also expressed in glial cells and in neuroendocrine bovine adrenal chromaffin and PC12 cells. NCS-1 is a regulatory protein involved in Ca^{2+}-dependent exocytosis of synaptic vesicles and dense core granules. NCS-1 also functions in the voltage-independent autocrine pathway that negatively regulates non-L-type Ca^{2+} channels.

REFERENCES

1. Pongs, O., et al. 1993. Frequenin—a novel calcium-binding protein that modulates synaptic efficacy in the *Drosophila* nervous system. Neuron 11: 15-28.
2. Cox, J.A., et al. 1994. Cation binding and conformational changes in VILIP and NCS-1, two neuron-specific calcium-binding proteins. J. Biol. Chem. 269: 32807-32813.
3. Olafsson, P., et al. 1997. The Ca^{2+} binding protein, frequenin is a nervous system-specific protein in mouse preferentially localized in neurites. Brain Res. Mol. Brain Res. 44: 73-82.
4. McFerran, B.W., et al. 1998. Neuronal Ca^{2+} sensor 1, the mammalian homologue of frequenin, is expressed in chromaffin and PC12 cells and regulates neurosecretion from dense-core granules. J. Biol. Chem. 273: 22768-22772.
5. Braunewell, K.H., et al. 1999. Intracellular neuronal calcium sensor proteins: a family of EF-hand calcium-binding proteins in search of a function. Cell Tissue Res. 295: 1-12.
6. Martone, M.E., et al. 1999. Cellular and subcellular distribution of the calcium-binding protein NCS-1 in the central nervous system of the rat. Cell Tissue Res. 295: 395-407.
7. Weiss, J.L., et al. 2000. NCS-1/Frequenin functions in an autocrine pathway regulating Ca^{2+} channels in bovine adrenal chromaffin cells. J. Biol. Chem. 275: 40082-40087.

CHROMOSOMAL LOCATION

Genetic locus: NCS1 (human) mapping to 9q34.11.

PRODUCT

NCS-1 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 transfections. Also see NCS-1 shRNA Plasmid (h): sc-36019-SH and NCS-1 shRNA (h) Lentiviral Particles: sc-36019-V as alternate gene silencing products.

For independent verification of NCS-1 (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-36019A, sc-36019B and sc-36019C.

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

NCS-1 siRNA (h) is recommended for the inhibition of NCS-1 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

NCS-1 (G-4): sc-376206 is recommended as a control antibody for monitoring of NCS-1 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 NCS-1 gene expression knockdown using RT-PCR Primer: NCS-1 (h)-PR: sc-36019-PR (20 µl, 508 bp). 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.