

4.1N siRNA (h): sc-105013

BACKGROUND

4.1N protein (band 4.1-like protein 1, neuronal protein 4.1) binds and stabilizes D2 and D3 dopamine receptors at the neuronal plasma membrane. 4.1 adapter proteins mediate interactions between the cytoskeleton and the overlying plasma membrane. These multiple 4.1N interactions with the cell cytoskeleton and plasma membrane may confer stability and plasticity to neuronal membrane. The 4.1N protein is expressed highly in the brain, and is found at lower levels in heart, kidney, pancreas, placenta, lung and skeletal muscle. Four homologous genes (4.1R, 4.1G, 4.1N, and 4.1B) undergo complex alternative splicing. The distribution of these 4.1 spliced gene products along the nephron suggests their involvement in targeting of selected transmembrane proteins in kidney epithelium and, therefore, in regulation of specific kidney functions.

REFERENCES

1. Ye, K., et al. 1999. Protein 4.1N binding to nuclear mitotic apparatus protein in PC12 cells mediates the antiproliferative actions of nerve growth factor. *J. Neurosci.* 19: 10747-10756.
2. Ye, K., et al. 2000. Pike. A nuclear gtpase that enhances PI3kinase activity and is regulated by protein 4.1N. *Cell* 103: 919-930.
3. Binda, A.V., et al. 2002. D2 and D3 dopamine receptor cell surface localization mediated by interaction with protein 4.1N. *Mol. Pharmacol.* 62: 507-513.
4. Ramez, M., et al. 2003. Distinct distribution of specific members of protein 4.1 gene family in the mouse nephron. *Kidney Int.* 63: 1321-1337.
5. Zhang, S., et al. 2003. Protein 4.1N is required for translocation of inositol 1,4,5-trisphosphate receptor type 1 to the basolateral membrane domain in polarized Madin-Darby canine kidney cells. *J. Biol. Chem.* 278: 4048-4056.
6. Fukatsu, K., et al. 2004. Lateral diffusion of inositol 1,4,5-trisphosphate receptor type 1 is regulated by actin filaments and 4.1N in neuronal dendrites. *J. Biol. Chem.* 279: 48976-48982.

CHROMOSOMAL LOCATION

Genetic locus: EPB41L1 (human) mapping to 20q11.23.

PRODUCT

4.1N 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see 4.1N shRNA Plasmid (h): sc-105013-SH and 4.1N shRNA (h) Lentiviral Particles: sc-105013-V as alternate gene silencing products.

For independent verification of 4.1N (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-105013A, sc-105013B and sc-105013C.

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 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

4.1N siRNA (h) is recommended for the inhibition of 4.1N 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 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.

GENE EXPRESSION MONITORING

4.1N (B-2): sc-374367 is recommended as a control antibody for monitoring of 4.1N 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 4.1N gene expression knockdown using RT-PCR Primer: 4.1N (h)-PR: sc-105013-PR (20 μ l). 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.