

4.1R siRNA (h): sc-40295

BACKGROUND

The 4.1 gene family encodes a group of multifunctional cytoskeletal proteins (4.1R, 4.1G, 4.1N and 4.1B), which are predominantly expressed in the nervous system. 4.1G is a protein that stabilizes spectrin-Actin interactions and is associated with hereditary elliptocytosis. Red blood cell 4.1, designated 4.1R, is a multifunctional protein that is essential for maintaining erythrocyte shape and membrane mechanical properties. Both 4.1R and 4.1G are distributed in a unique pattern in the cerebellum and are believed to modulate the membrane mechanical properties of neuronal cells by promoting fodrin/actin association. 4.1N and 4.1B, designated EPB41L1 and EPB41L3, respectively, are strongly expressed in the brain. Antibodies to 4.1N have been reported to detect multiple forms, each enriched in postsynaptic density preparations relative to brain homogenate. Antibodies to 4.1B have been reported to detect two forms.

REFERENCES

1. Peters, L.L., et al. 1998. Four paralogous protein 4.1 genes map to distinct chromosomes in mouse and human. *Genomics* 54: 348-350.
2. Takakuwa, Y. 2000. Protein 4.1, a multifunctional protein of the erythrocyte membrane skeleton: structure and functions in erythrocytes and nonerythrocyte cells. *Int. J. Hematol.* 72: 298-309.
3. Ohara, R., et al. 2000. Type II brain 4.1 (4.1B/KIAA0987), a member of the protein 4.1 family, is localized to neuronal paranodes. *Brain Res. Mol. Brain Res.* 85: 41-52.
4. Kontogianni-Konstantopoulos, A., et al. 2001. The prototypical 4.1R-10-kDa domain and the 4.1G-10-kDa paralog mediate fodrin-Actin complex formation. *J. Biol. Chem.* 276: 20679-20687.
5. Scott, C., et al. 2001. Protein 4.1 in forebrain postsynaptic density preparations: enrichment of 4.1 gene products and detection of 4.1R binding proteins. *Eur. J. Biochem.* 268: 1084-1094.
6. LocusLink Report (LocusID: 2036). <http://www.ncbi.nlm.nih.gov/LocusLink/>

CHROMOSOMAL LOCATION

Genetic locus: EPB41 (human) mapping to 1p35.3.

PRODUCT

4.1R 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 4.1R shRNA Plasmid (h): sc-40295-SH and 4.1R shRNA (h) Lentiviral Particles: sc-40295-V as alternate gene silencing products.

For independent verification of 4.1R (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-40295A, sc-40295B and sc-40295C.

PROTOCOLS

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

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

4.1R siRNA (h) is recommended for the inhibition of 4.1R 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

4.1R (B-11): sc-166759 is recommended as a control antibody for monitoring of 4.1R 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 4.1R gene expression knockdown using RT-PCR Primer: 4.1R (h)-PR: sc-40295-PR (20 μ l). 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.