

CASPR siRNA (h): sc-41915

BACKGROUND

Neurexins comprise a family of neuronal cell surface proteins, which include neurexin I (NRXN1), neurexin II (NRXN2), neurexin III (NRXN3) and CASPR (neurexin IV). Neurexins I-III are expressed as α and β isoforms. The α isoforms are made of three cassettes, which contain two LNS (laminin A, neurexins, sex hormone-binding)-domains separated by EGF domains, followed by a transmembrane region and a 55 amino acid cytoplasmic C-terminal. The α isoforms bind to neuroligins at the second LNS site, and to the excitatory neurotoxin α -latrotoxin. The β isoforms have only one LNS-domain, bind to neuroligins and play a role in the formation and remodeling of synapses. CASPR (for contactin-associated protein 1, also designated paranodin in mouse), contains an extracellular domain similar to the other three neurexins, and binds to the surface glycoprotein contactin. CASPR and the closely related CASPR2, a mammalian homolog of *Drosophila* neurexin IV (Nrx-IV), demarcate distinct subdomains in myelinated axons. Specifically, CASPR exists at the paranodal junctions, while CASPR2 co-localizes with Shaker-like K⁺ channels in the juxtaparanodal region. CASPR may play a role in the communication of glial cells and neurons during development.

REFERENCES

1. Ichtchenko, K., et al. 1996. Structures, alternative splicing, and neurexin binding of multiple neuroligins. *J. Biol. Chem.* 271: 2676-2682.
2. Einheber, S., et al. 1997. The axonal membrane protein CASPR, a homologue of neurexin IV, is a component of the septate-like paranodal junctions that assemble during myelination. *J. Cell Biol.* 139: 1495-1506.
3. Nguyen, T., et al. 1997. Binding properties of neuroligin 1, and Neurexin I β reveal function as heterophilic cell adhesion molecules. *J. Biol. Chem.* 272: 26032-26039.
4. Peles, E., et al. 1997. Identification of a novel contactin-associated transmembrane receptor with multiple domains implicated in protein-protein interactions. *EMBO J.* 16: 978-988.
5. Poliak, S., et al. 1997. CASPR2, a new member of the neurexin superfamily, is localized at the juxtaparanodes of myelinated axons and associates with K⁺ channels. *Neuron* 24: 1037-1104.
6. Missler, M., et al. 1998. Neurexophilin binding to β -neurexins. *J. Biol. Chem.* 273: 34716-34723.

CHROMOSOMAL LOCATION

Genetic locus: CNTNAP1 (human) mapping to 17q21.2.

PRODUCT

CASPR 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 CASPR shRNA Plasmid (h): sc-41915-SH and CASPR shRNA (h) Lentiviral Particles: sc-41915-V as alternate gene silencing products.

For independent verification of CASPR (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-41915A, sc-41915B and sc-41915C.

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

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

CASPR (A-3): sc-374489 is recommended as a control antibody for monitoring of CASPR 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 CASPR gene expression knockdown using RT-PCR Primer: CASPR (h)-PR: sc-41915-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.

PROTOCOLS

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