

SEMA4G siRNA (h): sc-62998

BACKGROUND

Semaphorins are a family of cell surface and secreted proteins that are conserved from insects to humans. Members of this family of proteins are approximately 750 amino acids in length (including signal sequences) and are defined by a conserved extracellular "semaphorin" domain of approximately 500 amino acids containing 14-16 cysteines, blocks of conserved sequences and no obvious repeats. Secreted and cell-bound semaphorins chemically attract and repel the growth of neural axons, guiding the development of intricate networks of neural tissue. SEMA4G (semaphorin-4G) is an 838 amino acid single-pass type I membrane protein that is thought to play a role in axon guidance. Existing as three alternatively spliced isoforms, SEMA4G contains one Ig-like C2-type (immunoglobulin-like) domain, a PSI domain and a single SEMA domain.

REFERENCES

1. Li, H., et al. 1999. Characterization and expression of sema4g, a novel member of the semaphorin gene family. *Mech. Dev.* 87: 169-173.
2. Holtmaat, A.J., et al. 2002. Semaphorins: contributors to structural stability of hippocampal networks? *Prog. Brain Res.* 138: 17-38.
3. Dickson, B.J. 2002. Molecular mechanisms of axon guidance. *Science* 298: 1959-1964.
4. Pasterkamp, R.J. and Kolodkin, A.L. 2003. Semaphorin junction: making tracks toward neural connectivity. *Curr. Opin. Neurobiol.* 13: 79-89.
5. Burkhardt, C., et al. 2005. Semaphorin 4B interacts with the post-synaptic density protein PSD-95/SAP90 and is recruited to synapses through a C-terminal PDZ-binding motif. *FEBS Lett.* 579: 3821-3828.
6. Shifman, M.I. and Selzer, M.E. 2006. Semaphorins and their receptors in lamprey CNS: Cloning, phylogenetic analysis, and developmental changes during metamorphosis. *J. Comp. Neurol.* 497: 115-132.

CHROMOSOMAL LOCATION

Genetic locus: SEMA4G (human) mapping to 10q24.31.

PRODUCT

SEMA4G 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 SEMA4G shRNA Plasmid (h): sc-62998-SH and SEMA4G shRNA (h) Lentiviral Particles: sc-62998-V as alternate gene silencing products.

For independent verification of SEMA4G (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-62998A, sc-62998B and sc-62998C.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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

SEMA4G siRNA (h) is recommended for the inhibition of SEMA4G 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 60 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

SEMA4G (N-18): sc-67951 is recommended as a control antibody for monitoring of SEMA4G 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor SEMA4G gene expression knockdown using RT-PCR Primer: SEMA4G (h)-PR: sc-62998-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.