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CD100 siRNA (h): sc-42817

BACKGROUND

Semaphorins comprise a family of phylogenetically conserved proteins containing a 500 amino acid region termed the "Sema domain" in the amino-terminus. The semaphorin family is subdivided into eight groups. CD100 (SEMA4D) is a transmembrane protein belonging to group IV of the semaphorin family. CD100 is expressed in both embryonic and adult kidney, heart, lung and neural tissues. Plexin-B1, a receptor expressed abundantly in fetal brain and kidney, binds CD100 with high affinity. CD100 is the only member of the semaphorin family that is expressed in the immune system. CD100 activates T cell proliferation and promotes B cell aggregation. CD100 regulates B cell response by negating an inhibitory signal from CD72 that normally diminishes B cell response. This signal is turned off when CD100 binds to CD72 and induces tyrosine dephosphorylation of CD72, followed by the dissociation of SHP-1 from CD72. The gene encoding human CD100 maps to chromosome 9q22.2.

REFERENCES

1. Bougeret, C., et al. 1992. Increased surface expression of a newly identified 150 kDa dimer early after human T lymphocyte activation. *J. Immunol.* 148: 318-323.
2. Herold, C., et al. 1995. Activation signals are delivered through two distinct epitopes of CD100, a unique 150 kDa human lymphocyte surface structure previously defined by BB18 mAb. *Int. Immunol.* 7: 1-8.
3. Furuyama, T., et al. 1996. Identification of a novel transmembrane semaphorin expressed on lymphocytes. *J. Biol. Chem.* 271: 33376-33381.
4. Hall, K.T., et al. 1996. Human CD100, a novel leukocyte semaphorin that promotes B-cell aggregation and differentiation. *Proc. Natl. Acad. Sci. USA* 93: 11780-11785.
5. Tamagnone, L., et al. 1999. Plexins are a large family of receptors for transmembrane, secreted, and GPI-anchored semaphorins in vertebrates. *Cell* 99: 71-80.

CHROMOSOMAL LOCATION

Genetic locus: SEMA4D (human) mapping to 9q22.2.

PRODUCT

CD100 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 CD100 shRNA Plasmid (h): sc-42817-SH and CD100 shRNA (h) Lentiviral Particles: sc-42817-V as alternate gene silencing products.

For independent verification of CD100 (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-42817A, sc-42817B and sc-42817C.

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

CD100 siRNA (h) is recommended for the inhibition of CD100 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

CD100 (C-3): sc-390675 is recommended as a control antibody for monitoring of CD100 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 CD100 gene expression knockdown using RT-PCR Primer: CD100 (h)-PR: sc-42817-PR (20 μ l, 568 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.