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Thrombospondin 4 siRNA (m): sc-43194

BACKGROUND

The Thrombospondin proteins (TSP 1-5) compose a family of glycoproteins that are involved in cell-to-cell and cell-to-matrix signaling. These extracellular, cell-surface proteins form complexes of both homo- and hetero-multimers. Thrombospondins play a role in development, aggregation of platelets, adhesion and migration of cells and progression of cells through the growth cycle. Thrombospondin 1 is released from platelets in response to Thrombin stimulation and is a transient component of the extracellular matrix of developing and repairing tissues. Thrombospondin 2 shares a high degree of homology with Thrombospondin 1, and is thought to have overlapping but unique functions. Thrombospondin 3 is a developmentally regulated heparin-binding protein. Thrombospondin 4 is neuronally expressed and stimulates neurite outgrowth.

REFERENCES

1. Mosher, D.F. 1990. Physiology of thrombospondin. *Annu. Rev. Med.* 41: 85-97.
2. Bornstein, P., et al. 1991. A second, expressed thrombospondin gene (Thbs2) exists in the mouse genome. *J. Biol. Chem.* 266: 12821-12824.
3. LaBell, T.L., et al. 1992. Thrombospondin II: partial cDNA sequence, chromosome location and expression of a second member of the thrombospondin gene family in humans. *Genomics* 12: 421-429.
4. O'Rourke, K.M., et al. 1992. Thrombospondin 1 and Thrombospondin 2 are expressed as both homo- and heterotrimers. *J. Biol. Chem.* 267: 24921-24924.
5. Jahav, J. 1993. The functions of thrombospondin and its involvement in physiology and pathophysiology. *Biochem. Biophys. Acta.* 1182: 1-14.
6. Arber, S., et al. 1995. Thrombospondin-4, an extracellular matrix protein expressed in the developing and adult nervous system promotes neurite outgrowth. *J. Cell Biol.* 131: 1083-1094.
7. Qabar, A., et al. 1995. Thrombospondin 3 is a pentameric molecule held together by interchain disulfide linkage involving two cysteine residues. *J. Biol. Chem.* 270: 12725-12729.

CHROMOSOMAL LOCATION

Genetic locus: Thbs4 (mouse) mapping to 13 C3.

PRODUCT

Thrombospondin 4 siRNA (m) 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 Thrombospondin 4 shRNA Plasmid (m): sc-43194-SH and Thrombospondin 4 shRNA (m) Lentiviral Particles: sc-43194-V as alternate gene silencing products.

For independent verification of Thrombospondin 4 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43194A, sc-43194B and sc-43194C.

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

Thrombospondin 4 siRNA (m) is recommended for the inhibition of Thrombospondin 4 expression in mouse 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

Thrombospondin 4 (F-7): sc-390734 is recommended as a control antibody for monitoring of Thrombospondin 4 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 Thrombospondin 4 gene expression knockdown using RT-PCR Primer: Thrombospondin 4 (m)-PR: sc-43194-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.