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PSGL-1 siRNA (m): sc-42833

BACKGROUND

PSGL-1 (P-Selectin glycoprotein ligand, also designated CD162) exists as a disulfide-linked homodimer. PSGL-1 is a type 1 membrane protein that localizes on the tips of microvilli of leukocytes. Its extracellular domain is rich in serines, threonines and prolines, and includes a series of 15 and 16 decameric repeats in HL-60 and U-937 cells and human leukocytes, respectively. Although PSGL-1 appears to be the sole receptor for P-Selectin on human hematopoietic cells, it also interacts with E-Selectin through a unique binding site. In order to bind PSGL-1 to either E-Selectin or P-Selectin, PSGL-1 must be sialylated and fucosylated. PSGL-1 is a mucin-like molecule, much like leukosialin (CD43), CD164 and CD34. These proteins belong to an emerging family of cell adhesion receptors called sialomucins, which transduce negative signals in hematopoietic cells.

REFERENCES

- Moore, K., et al. 1992. Identification of a specific glycoprotein ligand for P-Selectin (CD62) on Myeloid cells. *J. Biol. Chem.* 118: 445-456.
- Sako, D., et al. 1993. Expression cloning of a functional glycoprotein ligand for P-Selectin. *Cell* 75: 1179-1186.
- Veldman, G., et al. 1995. Genomic organization and chromosomal localization of the gene encoding human P-Selectin glycoprotein ligand. *J. Biol. Chem.* 270: 16470-16475.
- Patel, K., et al. 1995. Neutrophils use both shared and distinct mechanisms to adhere to selectins under static and flow conditions. *J. Clin. Invest.* 96: 1887-1896.
- Li, F., et al. 1996. Visualization of P-Selectin glycoprotein ligand-1 as a highly extended molecule and mapping of protein epitopes for monoclonal antibodies. *J. Biol. Chem.* 271: 6342-6348.

CHROMOSOMAL LOCATION

Genetic locus: Selpl (mouse) mapping to 5 F.

PRODUCT

PSGL-1 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 PSGL-1 shRNA Plasmid (m): sc-42833-SH and PSGL-1 shRNA (m) Lentiviral Particles: sc-42833-V as alternate gene silencing products.

For independent verification of PSGL-1 (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-42833A, sc-42833B and sc-42833C.

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.

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

PSGL-1 siRNA (m) is recommended for the inhibition of PSGL-1 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

PSGL-1 (PL2): sc-18856 is recommended as a control antibody for monitoring of PSGL-1 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 PSGL-1 gene expression knockdown using RT-PCR Primer: PSGL-1 (m)-PR: sc-42833-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.