

Endoglycan siRNA (h): sc-72322

BACKGROUND

Endoglycan, also known as podocalyxin-like 2, exists as a disulfide-dependent homodimer and belongs to the CD34 family of proteins. It is a proteoglycan modified with chondroitin sulfate chains and (with appropriate post-translational modifications) functions as an L-selectin ligand, interacting via sulfation on two tyrosine residues. Endoglycan contains a membrane proximal globular domain, a single pass transmembrane domain, a mucin-like domain and a highly acidic N-terminal with O-linked sLe^x structures. It is widely expressed with distribution including endothelial cells, hematopoietic precursors and leukocyte subpopulations. Endoglycan is similar to PSGL-1 in that it also exhibits catch-slip transitional bonds. These bonds help to mediate the rolling and tethering of circulating leukocytes on vascular surfaces, typically during immune surveillance and inflammation. The ligand activity of Endoglycan is dependent on sialylation and fucosylation and may mediate adhesion events.

REFERENCES

1. Sassetti, C., et al. 2000. Identification of Endoglycan, a member of the CD34/podocalyxin family of sialomucins. *J. Biol. Chem.* 275: 9001-9010.
2. Simmons, P.J., et al. 2001. Mucin-like molecules as modulators of the survival and proliferation of primitive hematopoietic cells. *Ann. N.Y. Acad. Sci. USA* 938: 196-206.
3. Fieger, C.B., et al. 2003. Endoglycan, a member of the CD34 family, functions as an L-Selectin ligand through modification with tyrosine sulfation and sialyl Lewis x. *J. Biol. Chem.* 278: 27390-27398.
4. Kerosuo, L., et al. 2004. Podocalyxin in human haematopoietic cells. *Br. J. Haematol.* 124: 809-818.
5. Sarangapani, K.K., et al. 2004. Low force decelerates L-Selectin dissociation from P-Selectin glycoprotein ligand-1 and endoglycan. *J. Biol. Chem.* 279: 2291-2298.
6. McEver, R.P. 2005. A sulfated address for lymphocyte homing. *Nat. Immunol.* 6: 1067-1069.
7. van der Zwaag, B., et al. 2005. Identifying new candidate genes for hereditary facial paresis on chromosome 3q21-q22 by RNA *in situ* hybridization in mouse. *Genomics* 86: 55-67.
8. Furness, S.G., et al. 2006. Beyond mere markers: functions for CD34 family of sialomucins in hematopoiesis. *Immunol. Res.* 34: 13-32.
9. Tan, P.C., et al. 2006. Na⁺/H⁺ exchanger regulatory factor-1 is a hematopoietic ligand for a subset of the CD34 family of stem cell surface proteins. *Stem Cells* 24: 1150-1161.

CHROMOSOMAL LOCATION

Genetic locus: PODXL2 (human) mapping to 3q21.3.

PROTOCOLS

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

PRODUCT

Endoglycan 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 Endoglycan shRNA Plasmid (h): sc-72322-SH and Endoglycan shRNA (h) Lentiviral Particles: sc-72322-V as alternate gene silencing products.

For independent verification of Endoglycan (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-72322A, sc-72322B and sc-72322C.

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

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

Endoglycan (Y122): sc-73935 is recommended as a control antibody for monitoring of Endoglycan 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Endoglycan gene expression knockdown using RT-PCR Primer: Endoglycan (h)-PR: sc-72322-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.