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ALCAM siRNA (m): sc-43024

BACKGROUND

Activated leukocyte cell adhesion molecule (ALCAM), expressed on activated leukocytes T cells, B cells and monocytes, is a member of the immunoglobulin superfamily (IgSF) and identified as a CD6 ligand. CD6 is a type I membrane protein in the scavenger receptor cysteine rich protein superfamily that acts in T cell adhesion and costimulation. ALCAM mediates thymocyte-thymic epithelial cell adhesion via homophilic (ALCAM-ALCAM) and heterophilic (ALCAM-CD6) interactions involving a previously undescribed protein-protein interaction between a member of the scavenger receptor cysteine rich protein superfamily and the immunoglobulin superfamily.

REFERENCES

1. Bowen, M.A., et al. 1995. Cloning, mapping, and characterization of activated leukocyte-cell adhesion molecule (ALCAM), a CD6 ligand. *J. Exp. Med.* 181: 2213-2220.
2. Skonier, J.E., et al. 1996. Recognition of diverse proteins by members of the immunoglobulin superfamily: delineation of the receptor binding site in the human CD6 ligand ALCAM. *Biochemistry* 35: 12287-12291.
3. Bowen, M.A., et al. 1997. Structure and chromosomal location of the human CD6 gene: detection of five human CD6 isoforms. *J. Immunol.* 158: 1149-1156.
4. Bowen, M.A., et al. 1997. Characterization of mouse ALCAM (CD166): the CD6-binding domain is conserved in different homologs and mediates cross-species binding. *Eur. J. Immunol.* 27: 1469-1478.
5. Cortes, F., et al. 1999. HCA, an immunoglobulin-like adhesion molecule present on the earliest human hematopoietic precursor cells, is also expressed by stromal cells in blood-forming tissues. *Blood* 93: 826-837.
6. Bowen, M.A., et al. 1999. Adhesion molecules, their receptors and their regulation: analysis of CD6-activated leukocyte cell adhesion molecule (ALCAM/CD166) interactions. *Transplant. Proc.* 31: 795-796.

CHROMOSOMAL LOCATION

Genetic locus: Alcam (mouse) mapping to 16 B5.

PRODUCT

ALCAM siRNA (m) is a pool of 2 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 ALCAM shRNA Plasmid (m): sc-43024-SH and ALCAM shRNA (m) Lentiviral Particles: sc-43024-V as alternate gene silencing products.

For independent verification of ALCAM (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-43024A and sc-43024B.

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

ALCAM siRNA (m) is recommended for the inhibition of ALCAM 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

ALCAM (B-6): sc-74558 is recommended as a control antibody for monitoring of ALCAM 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 ALCAM gene expression knockdown using RT-PCR Primer: ALCAM (m)-PR: sc-43024-PR (20 μ l, 419 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.