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CERCAM siRNA (m): sc-142294

BACKGROUND

CERCAM (cerebral endothelial cell adhesion molecule), also known as GLT25D3 (glycosyltransferase 25 family member 3), is a 595 amino acid protein that is localized to the endoplasmic reticulum. Despite belonging to the glycosyltransferase 25 family, CERCAM has no glucosyltransferase activity and very low, if any, β -galactosyltransferase activity. CERCAM is highly expressed in secretory and nervous tissues and likely functions as a cell adhesion protein that assists leukocyte transmigration across the blood-brain barrier. There are two isoforms of CERCAM that are produced as a result of alternative splicing events.

REFERENCES

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2. Steeber, D.A., et al. 2000. Adhesion molecule cascades direct lymphocyte recirculation and leukocyte migration during inflammation. *Immunol. Res.* 22: 299-317.
3. Starzyk, R.M., et al. 2000. Cerebral cell adhesion molecule: a novel leukocyte adhesion determinant on blood-brain barrier capillary endothelium. *J. Infect. Dis.* 181: 181-187.
4. Muller, W.A. 2003. Leukocyte-endothelial-cell interactions in leukocyte transmigration and the inflammatory response. *Trends Immunol.* 24: 327-334.
5. Schegg, B., et al. 2009. Core glycosylation of collagen is initiated by two β (1-0)galactosyltransferases. *Mol. Cell. Biol.* 29: 943-952.
6. Greenwood, J., et al. 2010. Leukocyte-endothelial cell crosstalk at the blood-brain barrier: A prerequisite for successful immune cell entry to the brain. *Neuropathol. Appl. Neurobiol.* 37: 24-39.

CHROMOSOMAL LOCATION

Genetic locus: Cercam (mouse) mapping to 2 B.

PRODUCT

CERCAM 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 CERCAM shRNA Plasmid (m): sc-142294-SH and CERCAM shRNA (m) Lentiviral Particles: sc-142294-V as alternate gene silencing products.

For independent verification of CERCAM (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-142294A, sc-142294B and sc-142294C.

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

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

CERCAM (D-4): sc-514083 is recommended as a control antibody for monitoring of CERCAM 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 CERCAM gene expression knockdown using RT-PCR Primer: CERCAM (m)-PR: sc-142294-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.