HCAM siRNA (m): sc-35534



The Power to Question

BACKGROUND

Cell adhesion molecules (CAMs) are a family of closely related, cell surface glycoproteins that are involved in cell-cell interactions and are thought to play an important role in embryogenesis and development. HCAM, also known as CD44, LHR, MDU2, MDU3, MIC4, Pgp1, HCELL, MUTCH-I or ECMR-III, is a 742 amino acid single-pass type I membrane protein that is involved in hematopoiesis, lymphocyte activation and tumor metastasis. Functioning as a receptor for hyaluronic acid (HA) and interacting with ligands such as osteopontin (OPN), HCAM mediates both cell-cell and cell-matrix interactions, thereby playing an essential role in cell adhesion and cell migration. HCAM contains one Link domain and, due to alternative splicing events, is expressed as multiple isoforms, some of which are designated CD44R, CDw44, CD44S, CD44H (hematopoietic) and CD44E (epithelial). While most of the HCAM splice varients are expressed in tissues throughout the body, one specific isoform, namely CD44H, is expressed at high levels in cancer tissue, suggesting an important role for the CD44H splice varient in tumor progression.

REFERENCES

- McVoy, L.A., et al. 2005. CD44 and annexin A2 mediate the C5a chemotactic cofactor function of the vitamin D binding protein. J. Immunol. 175: 4754-4760.
- 2. Hanley, W.D., et al. 2006. Variant isoforms of CD44 are P- and L-selectin ligands on colon carcinoma cells. FASEB J. 20: 337-339.
- Sugahara, K.N., et al. 2006. Tumor cells enhance their own CD44 cleavage and motility by generating hyaluronan fragments. J. Biol. Chem. 281: 5861-5868.

CHROMOSOMAL LOCATION

Genetic locus: Cd44 (mouse) mapping to 2 E2.

PRODUCT

HCAM 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HCAM shRNA Plasmid (m): sc-35534-SH and HCAM shRNA (m) Lentiviral Particles: sc-35534-V as alternate gene silencing products.

For independent verification of HCAM (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-35534A, sc-35534B and sc-35534C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

 $\ensuremath{\mathsf{HCAM}}$ siRNA (m) is recommended for the inhibition of HCAM 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 µM in 66 µl. 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

HCAM (DF1485): sc-7297 is recommended as a control antibody for monitoring of HCAM 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 HCAM gene expression knockdown using RT-PCR Primer: HCAM (m)-PR: sc-35534-PR (20 μ l, 470 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Jong, A., et al. 2012. Hyaluronic acid receptor CD44 deficiency is associated with decreased *Cryptococcus neoformans* brain infection. J. Biol. Chem. 287: 15298-15306.
- Liu, Y., et al. 2014. Mesenchymal stem cells inhibit lipopolysaccharideinduced inflammatory responses of BV2 microglial cells through TSG-6. J. Neuroinflammation 11: 135.

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.

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