SANTA CRUZ BIOTECHNOLOGY, INC.

TEM1 siRNA (m): sc-61660

BACKGROUND

Tumor endothelial marker 1 (TEM1/endosialin) is a heavily glycosylated, type I transmembrane C-type lectin-like receptor of the Ras superfamily expressed in the vascular endothelium and on fibroblast-like cells in developing organs. Expression of TEM1 largely disappears in adulthood. TEM1 is structurally related to thrombomodulin and complement receptor C1qRp. It consists of three EGF-like domains, a C-type lectin domain and a Sushi domain. TEM1 is highly upregulated in tumor endothelium and is known to function in tumor growth and progression. For this reason TEM1 is a major target in anti-angiogenic tumor therapy. TEM1 may be responsible for concentrating liposomes on the surface of target cells and promoting their fusion with the cell membrane.

REFERENCES

1. Carson-Walter, E.B., et al. 2001. Cell surface tumor endothelial markers are conserved in mice and humans. Cancer Res. 61: 6649-6655.
2. Christian, S., et al. 2001. Molecular cloning and characterization of endosialin, a C-type lectin-like cell surface receptor of tumor endothelium. J. Biol. Chem. 276: 7408-7414.
3. Opavsky, R., et al. 2001. Molecular characterization of the mouse Tem1/endosialin gene regulated by cell density *in vitro* and expressed in normal tissues *in vivo*. J. Biol. Chem. 276: 38795-38807.
4. Brady, J2004. Human endosialin (tumor endothelial marker 1) is abundantly expressed in highly malignant and invasive brain tumors. J. Neuropathol. Exp. Neurol. 63: 1274-1283.
5. MacFadyen, J.R., et al. 2005. Endosialin (TEM1, CD248) is a marker of stromal fibroblasts and is not selectively expressed on tumour endothelium. FEBS Lett. 579: 2569-2575.
6. MacFadyen, J., et al. 2006. Endosialin is expressed on stromal fibroblasts and CNS pericytes in mouse embryos and is downregulated during development. Gene Expr. Patterns 7: 363-369.
7. Rupp, C., et al. 2006. Mouse endosialin, a C-type lectin-like cell surface receptor: expression during embryonic development and induction in experimental cancer neoangiogenesis. Cancer Immun. 6: 10.

CHROMOSOMAL LOCATION

Genetic locus: Cd248 (mouse) mapping to 19 A.

PRODUCT

TEM1 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 TEM1 shRNA Plasmid (m): sc-61660-SH and TEM1 shRNA (m) Lentiviral Particles: sc-61660-V as alternate gene silencing products.

For independent verification of TEM1 (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-61660A, sc-61660B and sc-61660C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

TEM1 siRNA (m) is recommended for the inhibition of TEM1 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

TEM1 (G-9): sc-377221 is recommended as a control antibody for monitoring of TEM1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 TEM1 gene expression knockdown using RT-PCR Primer: TEM1 (m)-PR: sc-61660-PR (20 µl). 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.

PROTOCOLS

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