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SMS1 siRNA (h): sc-44428

BACKGROUND

The SMS (sphingomyelin synthase) family is a group of integral membrane proteins that includes SMS1 (sphingomyelin synthase 1) and SMS2 (sphingomyelin synthase 2). SMS1 is located in the Golgi apparatus, whereas SMS2 resides primarily at the plasma membrane. Both are bidirectional lipid choline-phosphotransferases which convert phosphatidylcholine (PC) and ceramide to sphingomyelin (SM) and diacylglycerol (DAG) and vice versa, the direction of which depends on the relative concentrations of ceramide and diacylglycerol as phosphocholine acceptors. Therefore, sphingomyelin synthases are thought to be involved in both cell death and survival. Tricyclodecan-9-yl-xanthogenate (D609), a selective tumor cytotoxic agent, inhibits SMS activity, contributing to tumor cell cytotoxicity. SMS proteins are expressed in liver, muscle, heart, brain, stomach and kidney. SMS1 is expressed as four alternatively spliced mRNAs (SMS1 α 1, SMS1 α 2, SMS1 β and SMS1 γ) that translate into three different proteins (SMS1 α , SMS1 β and SMS1 γ), which differ in their tissue distribution and function.

REFERENCES

- Luberto, C. and Hannun, Y.A. 1998. Sphingomyelin synthase, a potential regulator of intracellular levels of ceramide and diacylglycerol during SV40 transformation. *J. Biol. Chem.* 273: 14550-14559.
- Huitema, K., van den Dikkenberg, J., Brouwers, J.F. and Holthuis, J.C. 2004. Identification of a family of animal sphingomyelin synthases. *EMBO J.* 23: 33-44.
- Yamaoka, S., Miyaji, M., Kitano, T., Umehara, H. and Okazaki, T. 2004. Expression cloning of a human cDNA restoring sphingomyelin synthesis and cell growth in sphingomyelin synthase-defective lymphoid cells. *J. Biol. Chem.* 279: 18688-18693.
- Meng, A., Luberto, C., Meier, P., Bai, A., Yang, X., Hannun, Y.A. and Zhou, D. 2004. Sphingomyelin synthase as a potential target for D609-induced apoptosis in U937 human monocytic leukemia cells. *Exp. Cell Res.* 292: 385-392.

CHROMOSOMAL LOCATION

Genetic locus: SGMS1 (human) mapping to 10q11.23.

PRODUCT

SMS1 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 SMS1 shRNA Plasmid (h): sc-44428-SH and SMS1 shRNA (h) Lentiviral Particles: sc-44428-V as alternate gene silencing products.

For independent verification of SMS1 (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-44428A, sc-44428B and sc-44428C.

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

TRB-3 siRNA (m) is recommended for the inhibition of TRB-3 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

SMS1 (G-8): sc-133135 is recommended as a control antibody for monitoring of SMS1 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 SMS1 gene expression knockdown using RT-PCR Primer: SMS1 (h)-PR: sc-44428-PR (20 μ l, 577 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.