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SMS2 siRNA (h): sc-44429

BACKGROUND

The sphingomyelin synthase (SMS) family is a group of integral membrane proteins that includes Sphingomyelin Synthase 1 (SMS1) and Sphingomyelin Synthase 2 (SMS2). Sphingomyelin Synthase 1 is located in the Golgi apparatus, whereas Sphingomyelin Synthase 2 resides primarily at the plasma membrane. Both are bidirectional lipid cholinephosphotransferases 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. Tricyclocodecan-9-yl-xanthogenate (D609), a selective tumor cytotoxic agent, inhibits sphingomyelin synthases, contributing to tumor cell cytotoxicity. Sphingomyelin synthases are expressed in liver, muscle, heart, brain, stomach and kidney. The Sphingomyelin Synthase 1 gene encodes four alternatively spliced mRNAs (SMS1 α 1, SMS1 α 2, SMS1 β and SMS1 γ) that translate into three different proteins (Sphingomyelin Synthase 1 α , Sphingomyelin Synthase 1 β and Sphingomyelin Synthase 1 γ), which differ in tissue distribution and function.

REFERENCES

1. Luberto, C., et al. 1998. Sphingomyelin Synthase, a potential regulator of intracellular levels of ceramide and diacylglycerol during SV40 transformation. *J. Biol. Chem.* 273: 14550-14559.
2. Huitema, K., et al. 2004. Identification of a family of animal sphingomyelin synthases. *EMBO J.* 23: 33-44.
3. Yamaoka, S., et al. 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.
4. Meng, A., et al. 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: SGMS2 (human) mapping to 4q25.

PRODUCT

SMS2 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 SMS2 shRNA Plasmid (h): sc-44429-SH and SMS2 shRNA (h) Lentiviral Particles: sc-44429-V as alternate gene silencing products.

For independent verification of SMS2 (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-44429A, sc-44429B and sc-44429C.

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

SMS2 siRNA (h) is recommended for the inhibition of SMS2 expression in human 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

SMS2 (7D10): sc-293384 is recommended as a control antibody for monitoring of SMS2 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 SMS2 gene expression knockdown using RT-PCR Primer: SMS2 (h)-PR: sc-44429-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.