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SANTA CRUZ BIOTECHNOLOGY, INC.

GD3 Synthase siRNA (m): sc-44587

BACKGROUND

GD3 Synthase (GD3S, SIAT8, ST8SiaI, ST8 α-N-acetyl-neuraminide α-2,8-sialyltransferase 1) is a type II membrane protein that catalyzes the transfer of sialic acid from CMP-sialic acid to GM3 to produce gangliosides GD3 and GT3. Gangliosides are membrane-bound glycosphingolipids containing sialic acid. Ganglioside GD3 is known to be important for cell adhesion and growth of cultured malignant cells. GD3 Synthase is found in the Golgi apparatus and is a member of glycosyltransferase family 29. GD3 Synthase can downregulate MMP-9 promoter activity in response to TNFα by association with NFκB and activation protein-1 (AP-1) sites in the MMP-9 promoter. GD3 Synthase has an apoptotic effect on ECV304 cells through downregulation of Bcl-2 expression via dephosphorylation of AKT and CREB.

REFERENCES

1. Martina, J.A., et al. 1998. Influence of N-glycosylation and N-glycan trimming on the activity and intracellular traffic of GD3 Synthase. J. Biol. Chem. 273: 3725-3731.
2. Kawai, H., et al. 1998. Embryonic stem cells with a disrupted GD3 Synthase gene undergo neuronal differentiation in the absence of b-series gangliosides. J. Biol. Chem. 273: 19634-19638.
3. Birkle, S., et al. 2000. Downregulation of GD3 ganglioside and its O-acetylated derivative by stable transfection with antisense vector against GD3 Synthase gene expression in hamster melanoma cells: effects on cellular growth, melanogenesis and dendricity. J. Neurochem. 74: 547-554.
4. Fukumoto, S., et al. 2000. GD3 Synthase gene expression in PC12 cells results in the continuous activation of TrkA and ERK 1/2 and enhanced proliferation. J. Biol. Chem. 275: 5832-5838.
5. Satake, H., et al. 2003. Genes modulated by expression of GD3 Synthase in Chinese hamster ovary cells. Evidence that the Tis21 gene is involved in the induction of GD3 9-O-acetylation. J. Biol. Chem. 278: 7942-7948.
6. Furukawa, K., et al. 2003. Isolation and functional analysis of the melanoma specific promoter region of human GD3 Synthase gene. Biochim. Biophys. Acta 1627: 71-78.

CHROMOSOMAL LOCATION

Genetic locus: St8sia1 (mouse) mapping to 6 G3.

PRODUCT

GD3 Synthase 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 GD3 Synthase shRNA Plasmid (m): sc-44587-SH and GD3 Synthase shRNA (m) Lentiviral Particles: sc-44587-V as alternate gene silencing products.

For independent verification of GD3 Synthase (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-44587A, sc-44587B and sc-44587C.

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

GD3 Synthase siRNA (m) is recommended for the inhibition of GD3 Synthase 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

GD3 Synthase (B-11): sc-390123 is recommended as a control antibody for monitoring of GD3 Synthase 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-IgGλ BP-HRP: sc-516132 or m-IgGλ BP-HRP (Cruz Marker): sc-516132-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-IgGλ BP-FITC: sc-516185 or m-IgGλ BP-PE: sc-516186 (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 GD3 Synthase gene expression knockdown using RT-PCR Primer: GD3 Synthase (m)-PR: sc-44587-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.