

β-1,4-GalNAc-T siRNA (h): sc-77837

BACKGROUND

The chondroitin N-acetylgalactosaminyltransferase family includes β-1,4-GalNAc-T, β-1,4-GalNAc-T2, β-1,4-GalNAc-T3 and β-1,4-GalNAc-T4. The β-1,4-GalNAc-T protein consists of a short N-terminal residue, a trans-membrane region and a long C-terminal residue, which includes a catalytic domain and localizes to the Golgi apparatus. β-1,4-GalNAc-T utilizes simple ganglioside GM3 as a substrate for more complex gangliosides GM2, GM1 and GD1a. β-1,4-GalNAc-T is expressed in normal brain tissues and in various malignant transformed cells, such as malignant melanoma, neuroblastoma and adult T cell leukemia. Mice lacking the β-1,4-GalNAc-T protein develop significant and progressive behavioral neuropathies, including deficits in reflexes, strength, coordination and balance. β-1,4-GalNAc-T is a potential molecular marker for detecting melanoma cells and monitoring tumor progression.

REFERENCES

1. Hidari, J.K., et al. 1994. β 1-4-N-acetylgalactosaminyltransferase can synthesize both asialoglycosphingolipid GM2 and glycosphingolipid GM2 *in vitro* and *in vivo*: isolation and characterization of a β 1-4-N-acetylgalactosaminyltransferase cDNA clone from rat ascites hepatoma cell line AH7974F. *Biochem. J.* 303: 957-965.
2. Lutz, M.S., et al. 1994. Cloned β 1,4 N-acetylgalactosaminyltransferase synthesizes GA2 as well as gangliosides GM2 and GD2. GM3 synthesis has priority over GA2 synthesis for utilization of lactosylceramide substrate *in vivo*. *J. Biol. Chem.* 269: 29227-29231.
3. Haraguchi, M., et al. 1995. The effects of the site-directed removal of N-glycosylation sites from β-1,4-N-acetylgalactosaminyltransferase on its function. *Biochem. J.* 312: 273-280.
4. Sango, K., et al. 1995. β1,4-N-acetylgalactosaminyltransferase involved in ganglioside synthesis: cDNA sequence, expression, and chromosome mapping of the mouse gene. *Genomics* 27: 362-365.
5. Furukawa, K., et al. 1996. Genomic organization and chromosomal assignment of the human β1, 4-N-acetylgalactosaminyltransferase gene. Identification of multiple transcription units. *J. Biol. Chem.* 271: 20836-20844.

CHROMOSOMAL LOCATION

Genetic locus: CSGALNACT1 (human) mapping to 8p21.3.

PRODUCT

β-1,4-GalNAc-T 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 μM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see β-1,4-GalNAc-T shRNA Plasmid (h): sc-77837-SH and β-1,4-GalNAc-T shRNA (h) Lentiviral Particles: sc-77837-V as alternate gene silencing products.

For independent verification of β-1,4-GalNAc-T (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-77837A, sc-77837B and sc-77837C.

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

β-1,4-GalNAc-T siRNA (h) is recommended for the inhibition of β-1,4-GalNAc-T 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor β-1,4-GalNAc-T gene expression knockdown using RT-PCR Primer: β-1,4-GalNAc-T (h)-PR: sc-77837-PR (20 μl, 543 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ji, X.Y., et al. 2016. Design and synthesis of cajanine analogues against hepatitis C virus through down-regulating host chondroitin sulfate N-acetylgalactosaminyltransferase 1. *J. Med. Chem.* 59: 10268-10284.

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.