

GalNAc-T9 siRNA (h): sc-75104

BACKGROUND

The UDP-N-acetyl- α -D-galactosamine:polypeptide N-acetylgalactosaminyltransferase (GalNAc-T) family of enzymes are substrate-specific proteins that catalyze the transfer of GalNAc (N-acetylgalactosamine) to serine and threonine residues onto various proteins, thereby initiating mucin-type O-linked glycosylation in the Golgi apparatus. GalNAc-T9 (polypeptide N-acetylgalactosaminyltransferase 9), also known as UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase 9, is a 603 amino acid single-pass type II membrane enzyme that catalyzes the O-glycosylation of in the brain. Its N-terminal domain is involved in substrate binding and manganese coordination, while the C-terminal domain is involved in UDP-Gal binding and catalytic reaction. GalNAc-T9 is specifically expressed in brain, where it is localized to frontal lobe, temporal lobe, putamen, spinal cord and cerebellum. It is weakly expressed in cerebral cortex. There are two isoforms of GalNAc-T9 that are produced as a result of alternative splicing events.

REFERENCES

1. Toba, S., Tenno, M., Konishi, M., Mikami, T., Itoh, N. and Kurosaka, A. 2000. Brain-specific expression of a novel human UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase (GalNAc-T9). *Biochim. Biophys. Acta* 1493: 264-268.
2. Ten Hagen, K.G., Bedi, G.S., Tetaert, D., Kingsley, P.D., Hagen, F.K., Balys, M.M., Beres, T.M., Degand, P. and Tabak, L.A. 2001. Cloning and characterization of a ninth member of the UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase family, ppGalNTase-T9. *J. Biol. Chem.* 276: 17395-17404.
3. Online Mendelian Inheritance in Man, OMIM[™]. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 606251. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Nelson, P.A., Sutcliffe, J.G. and Thomas, E.A. 2002. A new UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase mRNA exhibits predominant expression in the hypothalamus, thalamus and amygdala of mouse forebrain. *Brain Res. Gene Expr. Patterns* 1: 95-99.
5. Zhang, Y., Iwasaki, H., Wang, H., Kudo, T., Kalka, T.B., Hennet, T., Kubota, T., Cheng, L., Inaba, N., Gotoh, M., Togayachi, A., Guo, J., Hisatomi, H., Nakajima, K., Nishihara, S., Nakamura, M., Marth, J.D. and Narimatsu, H. 2003. Cloning and characterization of a new human UDP-N-acetyl- α -D-galactosamine:polypeptide N-acetylgalactosaminyltransferase, designated pp-GalNAc-T13, that is specifically expressed in neurons and synthesizes GalNAc α -serine/threonine antigen. *J. Biol. Chem.* 278: 573-584.

CHROMOSOMAL LOCATION

Genetic locus: GALNT9 (human) mapping to 12q24.33.

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.

PRODUCT

GalNAc-T9 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 GalNAc-T9 shRNA Plasmid (h): sc-75104-SH and GalNAc-T9 shRNA (h) Lentiviral Particles: sc-75104-V as alternate gene silencing products.

For independent verification of GalNAc-T9 (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-75104A, sc-75104B and sc-75104C.

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

GalNAc-T9 siRNA (h) is recommended for the inhibition of GalNAc-T9 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GalNAc-T9 gene expression knockdown using RT-PCR Primer: GalNAc-T9 (h)-PR: sc-75104-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.