

GalNAc-T5 siRNA (h): sc-75098

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-T5 (polypeptide N-acetylgalactosaminyltransferase 5), also known as UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferase 5, is a 940 amino acid protein that displays enzymatic activity toward EA2 peptide substrate with weaker activity toward Muc2 or Muc 1b substrates. 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. EXT2 directly interacts with GalNAc-T5, suggesting that these proteins may corroborate in glycosaminoglycan synthesis.

REFERENCES

1. Elhammer, A.P., et al. 1999. The acceptor specificity of UDP-GalNAc: polypeptide N-acetylgalactosaminyltransferases. *Glycoconj. J.* 16: 171-180.
2. Simmons, A.D., et al. 1999. A direct interaction between EXT proteins and glycosyltransferases is defective in hereditary multiple exostoses. *Hum. Mol. Genet.* 8: 2155-2164.
3. McCormick, C., et al. 2000. The putative tumor suppressors EXT1 and EXT2 form a stable complex that accumulates in the Golgi apparatus and catalyzes the synthesis of heparan sulfate. *Proc. Natl. Acad. Sci. USA* 97: 668-673.
4. Ten Hagen, K.G., et al. 2003. All in the family: the UDP-GalNAc:polypeptide N-acetylgalactosaminyltransferases. *Glycobiology* 13: 1R-16R.
5. Sjöblom, T., et al. 2006. The consensus coding sequences of human breast and colorectal cancers. *Science* 314: 268-274.
6. Tarp, M.A., et al. 2008. Mucin-type O-glycosylation and its potential use in drug and vaccine development. *Biochim. Biophys. Acta* 1780: 546-563.

CHROMOSOMAL LOCATION

Genetic locus: GALNT5 (human) mapping to 2q24.1.

PRODUCT

GalNAc-T5 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-T5 shRNA Plasmid (h): sc-75098-SH and GalNAc-T5 shRNA (h) Lentiviral Particles: sc-75098-V as alternate gene silencing products.

For independent verification of GalNAc-T5 (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-75098A, sc-75098B and sc-75098C.

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

GalNAc-T5 siRNA (h) is recommended for the inhibition of GalNAc-T5 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-T5 gene expression knockdown using RT-PCR Primer: GalNAc-T5 (h)-PR: sc-75098-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.