

GalNAc-T3 siRNA (h): sc-75096

BACKGROUND

The UDP-N-acetyl- α -D-galactosamine:polypeptide N-acetylgalactosaminyl-transferase (GalNAc-T) family of enzymes are substrate-specific proteins that catalyze the transfer of GalNAc (N-acetylgalactosaminyl) to serine and threonine residues onto various proteins, thereby initiating mucin-type O-linked glycosylation in the Golgi apparatus. GalNAc-T3, also known as GALNT3, HHS or HFTC, is a 633 amino acid single-pass type II membrane protein that localizes to the Golgi and contains one ricin B-type lectin domain. Expressed at high levels in kidney, testis, skin and pancreas, GalNAc-T3 uses manganese and calcium as cofactors to catalyze the first reaction in O-linked oligosaccharide biosynthesis, namely the transfer of an N-acetyl-D-galactosamine residue to a serine or threonine residue on a target protein. Defects in the gene encoding GalNAc-T3 are the cause of hyperphosphatemic familial tumoral calcinosis (HFTC) and hyperostosis-hyperphosphatemia syndrome (HHS). GalNAc-T3 is overexpressed in carcinoma tissue, suggesting a role in tumor development and metastasis.

REFERENCES

1. Bennett, E.P., et al. 1996. cDNA cloning and expression of a novel human UDP-N-acetyl- α -D-galactosamine: Polypeptide N-acetylgalactosaminyl-transferase, GalNAc-t3. *J. Biol. Chem.* 271: 17006-17012.
2. Gu, C., et al. 2004. Low expression of polypeptide GalNAc N-acetylgalactosaminyl transferase-3 in lung adenocarcinoma: impact on poor prognosis and early recurrence. *Br. J. Cancer.* 90: 436-442.
3. Topaz, O., et al. 2004. Mutations in GALNT3, encoding a protein involved in O-linked glycosylation, cause familial tumoral calcinosis. *Nat. Genet.* 36: 579-581.
4. Kato, K., et al. 2006. Polypeptide GalNAc-transferase T3 and familial tumoral calcinosis. Secretion of fibroblast growth factor 23 requires O-glycosylation. *J. Biol. Chem.* 281: 18370-18377.
5. Garringer, H.J., et al. 2007. Two novel GALNT3 mutations in familial tumoral calcinosis. *Am. J. Med. Genet. A* 143A: 2390-2396.

CHROMOSOMAL LOCATION

Genetic locus: GALNT3 (human) mapping to 2q24.3.

PRODUCT

GalNAc-T3 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-T3 shRNA Plasmid (h): sc-75096-SH and GalNAc-T3 shRNA (h) Lentiviral Particles: sc-75096-V as alternate gene silencing products.

For independent verification of GalNAc-T3 (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-75096A, sc-75096B and sc-75096C.

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-T3 siRNA (h) is recommended for the inhibition of GalNAc-T3 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-T3 gene expression knockdown using RT-PCR Primer: GalNAc-T3 (h)-PR: sc-75096-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.