

C4ST-1 siRNA (h): sc-60303

BACKGROUND

Sulfotransferase enzymes catalyze the sulfate conjugation of many hormones, neurotransmitters, drugs, and xenobiotic compounds. These cytosolic enzymes differ in their tissue distributions and substrate specificities, although the gene structure (number and length of exons) is similar among family members. Sulfotransferases are primarily expressed in liver and adrenal tissues where they add sulfate to steroids and bile acids. C4ST-1 (chondroitin 4-sulphotransferase-1) transfers sulfate from PAPS (adenosine 3'-phosphate 5'-phosphosulphate) to position 4-O of N-acetylgalactosamine in chondroitin. This sulfation is required for proper chondroitin sulfate localization, modulation of distinct signaling pathways, and cartilage growth plate morphogenesis. N-linked oligosaccharides attached to C4ST-1 contribute to the production and stability of the active form of C4ST-1.

REFERENCES

1. Hiraoka, N., et al. 2000. Molecular cloning and expression of two distinct human chondroitin 4-O-sulfotransferases that belong to the HNK-1 sulfotransferase gene family. *J. Biol. Chem.* 275: 20188-20196.
2. Xia, G., et al. 2000. Molecular cloning and expression of the pituitary glycoprotein hormone N-acetylgalactosamine-4-O-sulfotransferase. *J. Biol. Chem.* 275: 38402-38409.
3. Mikami, T., et al. 2003. Specificities of three distinct human chondroitin/dermatan N-acetylgalactosamine-4-O-sulfotransferases demonstrated using partially desulfated dermatan sulfate as an acceptor: implication of differential roles in dermatan sulfate biosynthesis. *J. Biol. Chem.* 278: 36115-36127.
4. Yamada, T., et al. 2004. Chondroitin 4-sulphotransferase-1 and chondroitin 6-sulphotransferase-1 are affected differently by uronic acid residues neighboring the acceptor GalNAc residues. *Biochem. J.* 384: 567-575.
5. Klüppel, M., et al. 2005. Maintenance of chondroitin sulfation balance by chondroitin 4-sulfotransferase-1 is required for chondrocyte development and growth factor signaling during cartilage morphogenesis. *Development* 132: 3989-4003.

CHROMOSOMAL LOCATION

Genetic locus: CHST11 (human) mapping to 12q23.3.

PRODUCT

C4ST-1 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 C4ST-1 shRNA Plasmid (h): sc-60303-SH and C4ST-1 shRNA (h) Lentiviral Particles: sc-60303-V as alternate gene silencing products.

For independent verification of C4ST-1 (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-60303A, sc-60303B and sc-60303C.

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

C4ST-1 siRNA (h) is recommended for the inhibition of C4ST-1 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.

GENE EXPRESSION MONITORING

C4ST-1 (L18): sc-100868 is recommended as a control antibody for monitoring of C4ST-1 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-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-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-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (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 C4ST-1 gene expression knockdown using RT-PCR Primer: C4ST-1 (h)-PR: sc-60303-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.