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SANTA CRUZ BIOTECHNOLOGY, INC.

ITI-H4 siRNA (h): sc-45402

BACKGROUND

The inter-α trypsin inhibitor (ITI) family is a group of structurally related plasma serine protease inhibitors synthesized in the liver and built up from different combinations of three highly homologous heavy chains (ITI-H1, ITI-H2 and ITI-H3) and one light chain (Bikunin). Another member of the ITI family, ITI-H4 (also known as I a IH4P) harbors a Pro-rich region (PRR) in its C-terminus. ITI is a glycoprotein composed of three polypeptides linked by chondroitin sulphate: two heavy chains, ITI-H1 and ITI-H2, and Bikunin. Bikunin confers the protease-inhibitor function of ITI. The heavy chains of the ITI family, designated as SHAPs (for serum-derived hyaluronan-associated proteins), bind covalently to hyaluronic acid (HA), resulting in pericellular matrix stabilization. While the ITI family is primarily composed of multi-polypeptide molecules, ITI-H4 is a single-chain molecule. And unlike other ITI family members, the gene transcriptions and products for rat and human ITIH4 demonstrate marked differences, suggesting possible species-specific functions for ITI-H4. The gene encoding human ITI-H4 maps to chromosome 3p21.1.

REFERENCES

1. Bourguignon, J., et al. 1993. Human pre-α-trypsin inhibitor-precursor heavy chain. cDNA and deduced amino-acid sequence. Eur. J. Biochem. 212: 771-776.
2. Sarafan, N., et al. 1995. The human inter-α-trypsin inhibitor genes respond differently to interleukin-6 in Hep G2 cells. Eur. J. Biochem. 227: 808-815.
3. Soury, E., et al. 1998. The H4P heavy chain of inter-α-inhibitor family largely differs in the structure and synthesis of its prolin-rich region from rat to human. Biochem. Biophys. Res. Commun. 243: 522-530.
4. Mizushima, S., et al. 1998. Gene expression of the two heavy chains and one light chain forming the inter-α-trypsin-inhibitor in human tissues. Biol. Pharm. Bull. 21: 167-169.
5. Bost, F., et al. 1998. Inter-α-trypsin inhibitor proteoglycan family—a group of proteins binding and stabilizing the extracellular matrix. Eur. J. Biochem. 252: 339-346.
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CHROMOSOMAL LOCATION

Genetic locus: ITIH4 (human) mapping to 3p21.1.

PRODUCT

ITI-H4 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 ITI-H4 shRNA Plasmid (h): sc-45402-SH and ITI-H4 shRNA (h) Lentiviral Particles: sc-45402-V as alternate gene silencing products.

For independent verification of ITI-H4 (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-45402A, sc-45402B and sc-45402C.

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

ITI-H4 siRNA (h) is recommended for the inhibition of ITI-H4 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.

GENE EXPRESSION MONITORING

ITI-H4 (F-9): sc-515353 is recommended as a control antibody for monitoring of ITI-H4 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 ITI-H4 gene expression knockdown using RT-PCR Primer: ITI-H4 (h)-PR: sc-45402-PR (20 µl). 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.