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ATIII siRNA (m): sc-44840

BACKGROUND

The serine proteinase inhibitors (serpins) compose a superfamily of proteins with a diverse set of functions, including the control of blood coagulation, complement activation, programmed cell death and development. Serpins are secreted glycoproteins that contain a stretch of peptide that mimics a true substrate for a corresponding serine protease. Antithrombin III (ATIII), an extracellular plasma protein, is a crucial serine protease inhibitor that regulates the coagulation cascade in blood. The inhibitory activity of ATIII is amplified in the presence of heparin. ATIII inhibits Thrombin and Factors IXa, Xa and XIa. Defects in the gene SERPINC1, which encodes for ATIII, can cause ATIII deficiency, an autosomal dominant disease which is a risk factor for hereditary thrombophilia.

REFERENCES

1. Mackie, M., et al. 1978. Familial thrombosis: inherited deficiency of antithrombin III. *Br. Med. J.* 1: 136-138.
2. Scully, M.F., et al. 1981. Hereditary antithrombin III deficiency in an English family. *Br. J. Haematol.* 47: 235-240.
3. Winter, J.H., et al. 1981. Transfusion studies in patients with familial antithrombin III (ATIII) deficiency: half-disappearance time of infused ATIII and influence of such infusion on platelet life-span. *Br. J. Haematol.* 49: 449-453.
4. Okajima, K., et al. 1995. ATIII Kumamoto II; a single mutation at Arg 393-His increased the affinity of ATIII for heparin. *Am. J. Hematol.* 48: 12-18.
5. Bayston, T.A., et al. 1999. Familial overexpression of β AT caused by an Asn 135-Thr substitution. *Blood* 93: 4242-4247.
6. Yeung, P.K. 2000. Transgenic ATIII (Genzyme). *IDrugs* 3: 669-673.
7. Niiya, K., et al. 2001. Two novel gene mutations in type I AT deficiency. *Int. J. Hematol.* 74: 469-472.

CHROMOSOMAL LOCATION

Genetic locus: Serpinc1 (mouse) mapping to 1 H2.1.

PRODUCT

ATIII siRNA (m) 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 ATIII shRNA Plasmid (m): sc-44840-SH and ATIII shRNA (m) Lentiviral Particles: sc-44840-V as alternate gene silencing products.

For independent verification of ATIII (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44840A, sc-44840B and sc-44840C.

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

ATIII siRNA (m) is recommended for the inhibition of ATIII expression in mouse 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

ATIII (H-7): sc-271987 is recommended as a control antibody for monitoring of ATIII 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 ATIII gene expression knockdown using RT-PCR Primer: ATIII (m)-PR: sc-44840-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.