SANTA CRUZ BIOTECHNOLOGY, INC.

TIMP-3 siRNA (m): sc-37023

BACKGROUND

TIMP-1, TIMP-2, TIMP-3 and TIMP-4 (for tissue inhibitor of metalloproteinases 1, 2, 3 and 4) complex with metalloproteinases such as collagenases, gelatinases and stromelysins, resulting in irreversible inactivation of the metalloproteinase. TIMP-1 has been found to be identical to EPA (erythroid-potentiation activity). Parathyroid hormone has been shown to be a regulator of TIMP-2 in osteoblastic cells. TIMP-3 may be involved in regulating trophoblastic invasion of the uterus and remodeling of the extracellular matrix during the folding of epithelia, and in the formation, branching and expansion of epithelial tubes. TIMP-4 is most highly expressed in heart, with low levels expressed in liver, brain, lung, thymus and spleen.

REFERENCES

1. Docherty, A.J., et al. 1985. Sequence of human tissue inhibitor of metalloproteinases and its identity to erythroid-potentiating activity. Nature 318: 66-69.
2. Carmichael, D.F., et al. 1986. Primary structure and cDNA cloning of human fibroblast collagenase inhibitor. Proc. Natl. Acad. Sci. USA 83: 2407-2411.
3. Cook, T.F., et al. 1994. Cloning and regulation of rat tissue inhibitor of metalloproteinase-2 in osteoblastic cells. Arch. Biochem. Biophys. 311: 313-320.
4. Silbiger, S.M., et al. 1994. Cloning of cDNAs encoding human TIMP-3, a novel member of the tissue inhibitor of metalloproteinase family. Gene 141: 293-297.
5. Apte, S.S., et al. 1994. Gene encoding a novel murine tissue inhibitor of metalloproteinases (TIMP), TIMP-3, is expressed in developing mouse epithelia, cartilage and muscle, and is located on mouse chromosome 10. Dev. Dyn. 200: 177-197.
6. Apte, S.S., et al. 1995. The gene structure of tissue inhibitor of metalloproteinases (TIMP)-3 and its inhibitory activities define the distinct TIMP gene family. J. Biol. Chem. 270: 14313-14318.
7. Greene, J., et al. 1996. Molecular cloning and characterization of human tissue inhibitor of metalloproteinase 4. J. Biol. Chem. 271: 30375-30380.

CHROMOSOMAL LOCATION

Genetic locus: Timp3 (mouse) mapping to 10 C1.

PRODUCT

TIMP-3 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see TIMP-3 shRNA Plasmid (m): sc-37023-SH and TIMP-3 shRNA (m) Lentiviral Particles: sc-37023-V as alternate gene silencing products.

For independent verification of TIMP-3 (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-37023A, sc-37023B and sc-37023C.

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

TIMP-3 siRNA (m) is recommended for the inhibition of TIMP-3 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 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

TIMP-3 (B-2): sc-373839 is recommended as a control antibody for monitoring of TIMP-3 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 TIMP-3 gene expression knockdown using RT-PCR Primer: TIMP-3 (m)-PR: sc-37023-PR (20 µl, 432 bp). 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.