SANTA CRUZ BIOTECHNOLOGY, INC.

COL18A1 siRNA (h): sc-43072

BACKGROUND

Collagen Type XV (COL15 gene product) and XVIII (COL18 gene product) form the new subgroup MULTIPLEXIN, within the diverse family of collagens, which contains nineteen distinct types of collagens found in vertebrates. Both Collagen Type XV and Collagen XVIII are characterized by extensive interruptions in their collagenous sequences. Members of the MULTIPLEXIN subgroup contain polypeptides with multiple triple-helical domains separated and flanked by non-triple-helical regions. Type XV is predominantly expressed in internal organs such as adrenal gland, kidney and pancreas. Type XVIII encodes two different α1 chains, which have different signal peptides and variant N-terminal non-collagenous NC1 domains of 495 and 303 amino acids. The long variant NC1-434 Type XVIII mRNAs are promi-nently expressed in liver, while the variant NC1-303 mRNAs are predominantly expressed in heart, kidney, placenta, prostate, ovary, skeletal muscle and small intestine. Endostatin is a fragment of the C-terminal domain NC1 of Collagen Type XV and Type XVIII that inhibits angiogenesis and tumor growth. Unlike endostatin-XVIII, endostatin-XV does not bind zinc or heparin, however both endostatins can inhibit chorioallantoic membrane angiogenesis induced by basic FGF or VEGF. Col-lagen Type XV and XVIII are also widely present in basement membrane zones, suggesting their roles in basement membrane-stromal interactions and in-volvement with angiogenic and pathological processes.

REFERENCES

1. Muragaki, Y., et al. 1994. The human α1(XV) Collagen chain contains a large amino-terminal non-triple helical domain with a tandem repeat structure and homology to α1 (XVIII) collagen. J. Biol. Chem. 269: 4042-4046.
2. Kivirikko, S., et al. 1994. Primary structure of α1 chain of human Type XV Collagen and exon-intron organization in the 3' region of the corresponding gene. J. Biol. Chem. 269: 4773-4779.
3. Oh, S.P., et al. 1994. Cloning of cDNA and genomic DNA encoding human Type XVIII Collagen and localization of the α1(XVIII) collagen gene to mouse chromosome 10 and human chromosome 21. Genomics 19: 494-499.
4. Myers, J.C., et al. 1997. Biochemical and immunohistochemical characterization of human Type XIX defines a novel class of basement membrane zone collagens. Am. J. Pathol. 151: 1729-1740.

CHROMOSOMAL LOCATION

Genetic locus: COL18A1 (human) mapping to 21q22.3.

PRODUCT

COL18A1 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 COL18A1 shRNA Plasmid (h): sc-43072-SH and COL18A1 shRNA (h) Lentiviral Particles: sc-43072-V as alternate gene silencing products.

For independent verification of COL18A1 (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-43072A, sc-43072B and sc-43072C.

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

COL18A1 siRNA (h) is recommended for the inhibition of COL18A1 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

COL18A1 (1837-46): sc-32720 is recommended as a control antibody for monitoring of COL18A1 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 COL18A1 gene expression knockdown using RT-PCR Primer: COL18A1 (h)-PR: sc-43072-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.