Mast Cell Tryptase siRNA (m): sc-44922



The Power to Ouestion

BACKGROUND

Mast Cells are connective tissue cells derived from blood-forming tissues that line arterial walls and secrete substances, which mediate inflammatory and immune responses. Mast Cell chymase, known as CMA1, is a major secreted serine protease that is involved in vasoactive peptide generation, extracellular matrix degradation and regulation of gland secretion. The human chymase gene, which maps to human chromosome 14q11.2, encodes a preproenzyme with a 19-amino acid signal peptide, an acidic 2-amino acid propeptide and a 226-amino acid catalytic domain. Tryptases comprise a family of trypsin-like serine proteases that are enzymatically active as heparin-stabilized tetramers. There are four functional genes for tryptase: α I, β I, β II and γ I, which map to human chromosome 16p13.3, with β tryptases representing the main isoenzymes expressed in mast cells. Mast Cell proteases are a family of rodent protein homologs to human tryptases that are specifically expressed in mast cells and may serve as highly specific markers in the analysis of Mast Cell heterogeneity, differentiation and function.

REFERENCES

- Huang, R.Y., et al. 1991. Cloning and structural analysis of MMCP-1, MMCP-4 and MMCP-5, three mouse mast cell-specific serine proteases. Eur. J. Immunol. 21: 1611-1621.
- 2. Caughey, G.H., et al. 1991. Structure, chromosomal assignment and deduced amino acid sequence of a human gene for mast cell chymase. J. Biol. Chem. 266: 12956-12963.
- Caughey, G.H., et al. 1993. The human mast cell chymase gene (CMA1): mapping to the cathepsin G/granzyme gene cluster and lineage-restricted expression. Genomics 15: 614-620.
- 4. Gurish, M.F., et al. 2001. The diverse roles of mast cells. J. Exp. Med. 194: 1-5.
- Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 118938. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
- 6. LocusLink Report (LocusID: 7176). http://www.ncbi.nlm.nih.gov/LocusLink/

CHROMOSOMAL LOCATION

Genetic locus: Tpsb2 (mouse) mapping to 17 A3.3.

PRODUCT

Mast Cell Tryptase 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 Mast Cell Tryptase shRNA Plasmid (m): sc-44922-SH and Mast Cell Tryptase shRNA (m) Lentiviral Particles: sc-44922-V as alternate gene silencing products.

For independent verification of Mast Cell Tryptase (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-44922A, sc-44922B and sc-44922C.

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

Mast Cell Tryptase siRNA (m) is recommended for the inhibition of Mast Cell Tryptase 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

Mast Cell Tryptase (AA1): sc-59587 is recommended as a control antibody for monitoring of Mast Cell Tryptase 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 Mast Cell Tryptase gene expression knockdown using RT-PCR Primer: Mast Cell Tryptase (m)-PR: sc-44922-PR (20 μ l, 549 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.

Santa Cruz Biotechnology, Inc. 1.800.457.3801 831.457.3801 Fax 831.457.3801 Europe +00800 4573 8000 49 6221 4503 0 www.scbt.com