

MC-CPA siRNA (h): sc-60994

BACKGROUND

Carboxypeptidase A (CPA) is a pancreatic exopeptidase which hydrolyses the peptide bond adjacent to the C-terminal end in polypeptide chains. Mast cell carboxypeptidase A (MC-CPA), a part of the peptidase M14 family, is a highly conserved metalloprotease localized to the secretory granules, along with tryptases and chymases. MC-CPA is stored as an active enzyme in the granule and is released, along with other inflammatory mediators, upon mast cell degranulation. MC-CPA mirrors pancreatic carboxypeptidase A in cleaving COOH-terminal aromatic and aliphatic amino acid residues. The optimum pH of MC-CPA is between neutral and basic, depending upon the substrate. The MC-CPA gene, CPA3, resides on chromosome 3q24 and contains 11 exons.

REFERENCES

1. Reynolds, D.S., et al. 1992. Cloning and characterization of the novel gene for MC-CPA. *J. Clin. Invest.* 89: 273-282.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 114851. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Henningsson, F., et al. 2003. Mast cell cathepsins C and S control levels of carboxypeptidase A and the chymase, mouse mast cell protease 5. *Biol. Chem.* 384: 1527-1531.
4. Welker, P., et al. 2004. Differential expression of mast cell characteristics in human myeloid cell lines. *Exp. Dermatol.* 13: 535-542.
5. Lundequist, A., et al. 2004. Cooperation between MC-CPA and the chymase mouse mast cell protease 4 in the formation and degradation of Angiotensin II. *J. Biol. Chem.* 279: 32339-32344.
6. Schwartz, L.B. 2005. Analysis of MC(T) and MC(TC) mast cells in tissue. *Methods Mol. Biol.* 315: 53-62.
7. Feyerabend, T.B., et al. 2005. Loss of histochemical identity in mast cells lacking carboxypeptidase A. *Mol. Cell. Biol.* 25: 6199-6210.
8. Henningsson, F., et al. 2005. A role for cathepsin E in the processing of mast-cell carboxypeptidase A. *J. Cell Sci.* 118: 2035-2042.
9. Jamur, M.C., et al. 2005. Identification and characterization of undifferentiated mast cells in mouse bone marrow. *Blood* 105: 4282-4289.

CHROMOSOMAL LOCATION

Genetic locus: CPA3 (human) mapping to 3q24.

PRODUCT

MC-CPA 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MC-CPA shRNA Plasmid (h): sc-60994-SH and MC-CPA shRNA (h) Lentiviral Particles: sc-60994-V as alternate gene silencing products.

For independent verification of MC-CPA (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-60994A, sc-60994B and sc-60994C.

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

MC-CPA siRNA (h) is recommended for the inhibition of MC-CPA 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 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor MC-CPA gene expression knockdown using RT-PCR Primer: MC-CPA (h)-PR: sc-60994-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.