SANTA CRUZ BIOTECHNOLOGY, INC.

Thimet oligopeptidase siRNA (h): sc-72152

BACKGROUND

Thimet oligopeptidase, also designated soluble metallo-endopeptidase, is a cytoplasmic protein belonging to the peptidase M3 family. The gene for the protein maps against chromosome 19q13.3. Thimet oligopeptidase can degrade the β-Amyloid precursor protein and generate amyloidogenic fragments. It is important in cytoplasmic peptide degradation and involved in metabolism of neuropeptides that are less than 20 amino acids in length. Thimet oligopeptidase is highly expressed in testis but can also be detected in liver, lung and kidney.

REFERENCES

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2. McKie, N., et al. 1993. Thimet oligopeptidase: similarity to "soluble angiotensin II-binding protein" and some corrections to the published amino acid sequence of the rat testis enzyme. Biochem. J. 295: 57-60.
3. McCool, S., et al. 2000. Expression of the thimet oligopeptidase gene is regulated by positively and negatively acting elements. DNA Cell Biol. 19: 729-738.
4. Edbauer, D., et al. 2002. Insulin-degrading enzyme rapidly removes the β-Amyloid precursor protein intracellular domain (AICD). J. Biol. Chem. 277: 13389-13393.
5. Oliveira, V., et al. 2002. Temperature and salts effects on the peptidase activities of the recombinant metallooligopeptidases neurolysin and thimet oligopeptidase. Eur. J. Biochem. 269: 4326-4334.
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7. Ray, K., et al. 2004. Crystal structure of human thimet oligopeptidase provides insight into substrate recognition, regulation, and localization. J. Biol. Chem. 279: 20480-20489.
8. Saric, T., et al. 2004. Pathway for degradation of peptides generated by proteasomes: a key role for Thimet oligopeptidase and other metallopeptidases. J. Biol. Chem. 279: 46723-46732.

CHROMOSOMAL LOCATION

Genetic locus: THOP1 (human) mapping to 19p13.3.

PRODUCT

Thimet oligopeptidase 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 Thimet oligopeptidase shRNA Plasmid (h): sc-72152-SH and Thimet oligopeptidase shRNA (h) Lentiviral Particles: sc-72152-V as alternate gene silencing products.

For independent verification of Thimet oligopeptidase (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-72152A, sc-72152B and sc-72152C.

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

Thimet oligopeptidase siRNA (h) is recommended for the inhibition of Thimet oligopeptidase 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

Thimet oligopeptidase (4D6): sc-53565 is recommended as a control antibody for monitoring of Thimet oligopeptidase 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 Thimet oligopeptidase gene expression knockdown using RT-PCR Primer: Thimet oligopeptidase (h)-PR: sc-72152-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.