

cathepsin L siRNA (h): sc-29938

BACKGROUND

The cathepsin family of proteolytic enzymes contains several diverse classes of proteases. The cysteine protease class comprises cathepsins B, L, H, K, S, and O. The aspartyl protease class is composed of cathepsins D and E. Cathepsin G is in the serine protease class. Most cathepsins are lysosomal and each is involved in cellular metabolism, participating in various events such as peptide biosynthesis and protein degradation. Cathepsin L (also designated major excreted protein, MEP or CATL) is a member of the peptidase C1 family and has been identified as a protein that is most closely related to cathepsin H. It is a lysosomal cysteine proteinase that mediates intracellular protein catabolism for collagen, elastin and α -1 protease inhibitor. Cathepsin L is a dimer composed of disulfide-linked heavy and light chains, both produced from a single protein precursor. At least two transcript variants encoding the same protein have been found for this gene. Transformed mouse fibroblasts stimulated by growth factors or tumor promoters secrete a form of cathepsin L.

REFERENCES

1. Ishidoh, K., et al. 1987. Molecular cloning and sequencing of cDNA for rat cathepsin L. *FEBS Lett.* 223: 69-73.
2. Joseph, L.J., et al. 1988. Complete nucleotide and deduced amino acid sequences of human and murine preprocathepsin L. An abundant transcript induced by transformation of fibroblasts. *J. Clin. Invest.* 81: 1621-1629.
3. Soderstrom, M., et al. 1999. Cathepsin expression during skeletal development. *Biochim. Biophys. Acta* 1446: 35-46.
4. Abudula, A., et al. 2001. Splice variants of human cathepsin L mRNA show different expression rates. *Biol. Chem.* 382: 1583-1591.

CHROMOSOMAL LOCATION

Genetic locus: CTSL (human) mapping to 9q21.33.

PRODUCT

cathepsin L 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 cathepsin L shRNA Plasmid (h): sc-29938-SH and cathepsin L shRNA (h) Lentiviral Particles: sc-29938-V as alternate gene silencing products.

For independent verification of cathepsin L (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-29938A, sc-29938B and sc-29938C.

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

cathepsin L siRNA (h) is recommended for the inhibition of cathepsin L 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.

GENE EXPRESSION MONITORING

cathepsin L (33/2): sc-32320 is recommended as a control antibody for monitoring of cathepsin L 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 cathepsin L gene expression knockdown using RT-PCR Primer: cathepsin L (h)-PR: sc-29938-PR (20 μ l, 510 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Calton, C.M., et al. 2013. Human papillomavirus type 16 does not require cathepsin L or B for infection. *J. Gen. Virol.* 94: 1865-1869.
2. Mao, Z., et al. 2019. CSN6 promotes the migration and invasion of cervical cancer cells by inhibiting autophagic degradation of cathepsin L. *Int. J. Biol. Sci.* 15: 1310-1324.
3. Wang, W., et al. 2019. Cathepsin L activated by mutant p53 and Egr-1 promotes ionizing radiation-induced EMT in human NSCLC. *J. Exp. Clin. Cancer Res.* 38: 61.
4. Ma, S., et al. 2021. Iron-dependent autophagic cell death induced by radiation in MDA-MB-231 breast cancer cells. *Front. Cell Dev. Biol.* 9: 723801.

RESEARCH USE

For research use only, not for use in diagnostic procedures.