

cathepsin A siRNA (h): sc-41469

BACKGROUND

The cathepsin family of proteolytic enzymes include several diverse classes of proteases. Cathepsins B, L, H, K, S and O comprise the cysteine protease class. Cathepsins D and E comprise the aspartyle protease class. The serine protease class includes cathepsin G. Cathepsins function in cellular metabolism and participate in peptide biosynthesis and protein degradation. Cathepsin A, a serine carboxypeptidase, exists in a high molecular weight lysosomal complex with β -galactosidase (β -gal) and α -neuraminidase (Neu1). Cathepsin A functions to protect β -gal and Neu1 from intralysosomal proteolysis. Deficiencies in cathepsin A lead to deficiencies in β -gal and Neu1. The gene encoding human cathepsin A maps to chromosome 20q13.12. Mutations in this gene cause galactosialidosis, a lysosomal storage disorder resulting from the β -gal and Neu1 deficiencies.

REFERENCES

1. Wiegant, J., et al. 1991. The gene encoding human protective protein (PPGB) is on chromosome 20. *Genomics* 10: 345-349.
2. Heusel, J.W., et al. 1993. Molecular cloning, chromosomal location, and tissue-specific expression of the murine cathepsin G gene. *Blood* 81: 1614-1623.
3. Shi, G.P., et al. 1995. Molecular cloning of human cathepsin O, a novel endoproteinase and homologue of rabbit OC2. *FEBS Lett.* 357: 129-134.
4. Tsukuba, T., et al. 2000. New functional aspects of cathepsin D and cathepsin E. *Mol. Cells* 10: 601-611.
5. Ostrowska, H., et al. 2003. Lysosomal high molecular weight multienzyme complex. *Cell. Mol. Biol. Lett.* 8: 19-24.

CHROMOSOMAL LOCATION

Genetic locus: CTSA (human) mapping to 20q13.12.

PRODUCT

cathepsin A 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 A shRNA Plasmid (h): sc-41469-SH and cathepsin A shRNA (h) Lentiviral Particles: sc-41469-V as alternate gene silencing products.

For independent verification of cathepsin A (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-41469A, sc-41469B and sc-41469C.

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 A siRNA (h) is recommended for the inhibition of cathepsin A 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 A (4526): sc-73766 is recommended as a control antibody for monitoring of cathepsin A 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 A gene expression knockdown using RT-PCR Primer: cathepsin A (h)-PR: sc-41469-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.