

# cystatin B siRNA (h): sc-44431

## BACKGROUND

Cystatin A (also designated STF1, STFA, stefin A or cystatin AS) and cystatin B (also designated PME, CST6, STFB, CPI-B, stefin B and liver thiol proteinase inhibitor) are thiol protease inhibitors that form complexes with papain and the cathepsins B, H, and L. Cystatin A, a cytoplasmic protein, is one of the precursor proteins of the cornified cell envelope in keratinocytes and plays a role in epidermal development and maintenance. Cystatin B protects against intracellular proteases leaking out of lysosomes and is primarily expressed in heart, liver and kidney.

## REFERENCES

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- Jerala, R., et al. 1988. Cloning a synthetic gene for human stefin B and its expression in *E. coli*. *FEBS Lett.* 239: 41-44.
- Pennacchio, L.A., et al. 1996. Mutations in the gene encoding cystatin B in progressive myoclonus epilepsy (EPM1). *Science* 271: 1731-1734.
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- Takahashi, H., et al. 1998. Structure and transcriptional regulation of the human cystatin A gene. The 12-O-tetradecanoylphorbol-13-acetate (TPA) responsive element-2 site (-272 to -278) on cystatin A gene is critical for TPA-dependent regulation. *J. Biol. Chem.* 273: 17375-17380.
- Takahashi, H., et al. 2001. Expression of human cystatin A by keratinocytes is positively regulated via the Ras/MEK1/MKK7/JNK signal transduction pathway but negatively regulated via the Ras/Raf-1/MEK1/ERK pathway. *J. Biol. Chem.* 276: 36632-36638.
- Jenko, S., et al. 2004. Different propensity to form amyloid fibrils by two homologous proteins—human stefins A and B: searching for an explanation. *Proteins* 55: 417-425.

## CHROMOSOMAL LOCATION

Genetic locus: CSTB (human) mapping to 21q22.3.

## PRODUCT

cystatin B 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see cystatin B shRNA Plasmid (h): sc-44431-SH and cystatin B shRNA (h) Lentiviral Particles: sc-44431-V as alternate gene silencing products.

For independent verification of cystatin B (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-44431A, sc-44431B and sc-44431C.

## RESEARCH USE

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

## 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

cystatin B siRNA (h) is recommended for the inhibition of cystatin B 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  $\mu$ M in 66  $\mu$ 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

cystatin B (F-5): sc-166561 is recommended as a control antibody for monitoring of cystatin B 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 $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor cystatin B gene expression knockdown using RT-PCR Primer: cystatin B (h)-PR: sc-44431-PR (20  $\mu$ l, 405 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.