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PCPE siRNA (h): sc-45728

BACKGROUND

Fibrillar collagen proteins are synthesized as procollagens that contain carboxyl- and amino-terminal peptide extensions (C- and N-propeptides). As procollagen is secreted from cells, these propeptides are cleaved and form mature helical fibrils. Procollagen C-endopeptidase enhancer 1 precursor (PCPE-1), also designated Type I procollagen COOH-terminal proteinase enhancer or PCOLCE, binds to the C-terminal propeptide of Type I procollagen. It is an extracellular matrix glycoprotein that can heighten the activity of procollagen C-proteinase in a substrate-specific way. PCPE-1 can greatly stimulate the action of tolloid metalloproteinases during procollagen processing. Expression of PCPE-1 has been shown to be highest in type I collagen-rich connective tissues such as skin and tendon.

REFERENCES

1. Takahara, K., et al. 1994. Type I procollagen COOH-terminal proteinase enhancer protein: identification, primary structure, and chromosomal localization of the cognate human gene (PCOLCE). *J. Biol. Chem.* 269: 26280-26285.
2. Scott, I.C., et al. 1999. Structural organization and expression patterns of the human and mouse genes for the type I procollagen COOH-terminal proteinase enhancer protein. *Genomics* 55: 229-234.
3. Mott, J.D., et al. 2000. Post-translational proteolytic processing of procollagen C-terminal proteinase enhancer releases a metalloproteinase inhibitor. *J. Biol. Chem.* 275: 1384-1390.
4. Baker, A.H., et al. 2002. Metalloproteinase inhibitors: biological actions and therapeutic opportunities. *J. Cell Sci.* 115: 3719-3727.
5. Ricard-Blum, S., et al. 2002. Interaction properties of the procollagen C-proteinase enhancer protein shed light on the mechanism of stimulation of BMP-1. *J. Biol. Chem.* 277: 33864-33869.
6. Bernocco, S., et al. 2003. Low resolution structure determination shows procollagen C-proteinase enhancer to be an elongated multidomain glycoprotein. *J. Biol. Chem.* 278: 7199-7205.

CHROMOSOMAL LOCATION

Genetic locus: PCOLCE (human) mapping to 7q22.1.

PRODUCT

PCPE-1 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 PCPE-1 shRNA Plasmid (h): sc-45728-SH and PCPE-1 shRNA (h) Lentiviral Particles: sc-45728-V as alternate gene silencing products.

For independent verification of PCPE-1 (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-45728A, sc-45728B and sc-45728C.

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 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

PCPE-1 siRNA (h) is recommended for the inhibition of PCPE-1 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

PCPE-1 (10D9): sc-73002 is recommended as a control antibody for monitoring of PCPE-1 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 PCPE-1 gene expression knockdown using RT-PCR Primer: PCPE-1 (h)-PR: sc-45728-PR (20 μ l). 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 for detailed protocols and support products.