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SANTA CRUZ BIOTECHNOLOGY, INC.

PCPE-1 shRNA (m) Lentiviral Particles: sc-45729-V

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 (mouse) mapping to 5 G2.

PRODUCT

PCPE-1 shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see PCPE-1 siRNA (m): sc-45729 and PCPE-1 shRNA Plasmid (m): sc-45729-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

PCPE-1 shRNA (m) Lentiviral Particles is recommended for the inhibition of PCPE-1 expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

PCPE-1 (K-18): sc-47315 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PCPE-1 gene expression knockdown using RT-PCR Primer: PCPE-1 (m)-PR: sc-45729-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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