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SANTA CRUZ BIOTECHNOLOGY, INC.

fibrillin-2 shRNA (m) Lentiviral Particles: sc-45972-V

BACKGROUND

Extracellular glycoproteins fibrillin-1 and -2 are major components of connective tissue microfibrils. Fibrillin-2 containing microfibrils regulate the early process of elastic fiber assembly in tissue. Mutations in the fibrillin-2 gene resulting in impaired assembly of fibrillin-2 may lead to molecular congenital contractural arachnodactyly. Fibrillin-2 constitutes the backbone of microfibrils which insert directly into the lamina densa of basement membranes. Epithelial cells primarily deposit fibrillin into the extracellular matrix in a nonfibrillar form. Mutations in the 8-cysteine motif of fibrillin-2 alters its binding to microfibril-associated glycoprotein-1 (MAGP-1), which may increase the severity of congenital contractural arachnodactyly.

REFERENCES

1. Mariencheck, M.C., et al. 1995. Fibrillin-1 and fibrillin-2 show temporal and tissue-specific regulation of expression in developing elastic tissues. Connect. Tissue Res. 31: 87-97.
2. Dzamba, B., et al. 2001. Assembly of epithelial cell fibrillins. J. Invest. Dermatol. 117: 1612-1620.
3. Lin, G., et al. 2002. Homo- and heterotypic fibrillin-1 and -2 interactions constitute the basis for the assembly of microfibrils. J. Biol. Chem. 277: 50795-804.
4. Quondamatteo, F., et al. 2002. Fibrillin-1 and fibrillin-2 in human embryonic and early fetal development. Matrix Biol. 21: 637-646.
5. Ritty, T.M., et al. 2003. Fibrillin-1 and -2 contain heparin-binding sites important for matrix deposition and that support cell attachment. Biochem. J. 375: 425-432.

CHROMOSOMAL LOCATION

Genetic locus: Fbn2 (mouse) mapping to 18 D3.

PRODUCT

fibrillin-2 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 fibrillin-2 siRNA (m): sc-45972 and fibrillin-2 shRNA Plasmid (m): sc-45972-SH as alternate gene silencing products.

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.

APPLICATIONS

fibrillin-2 shRNA (m) Lentiviral Particles is recommended for the inhibition of fibrillin-2 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

fibrillin-2 (H-10): sc-393968 is recommended as a control antibody for monitoring of fibrillin-2 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 goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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 goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor fibrillin-2 gene expression knockdown using RT-PCR Primer: fibrillin-2 (m)-PR: sc-45972-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.

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.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.