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LTBP-2 shRNA (h) Lentiviral Particles: sc-43388-V

BACKGROUND

Transforming growth factor (TGF) β is secreted as a part of an inactive complex that frequently contains latent TGF β -binding protein (LTBP). The LTBP family of proteins exhibit a multidomain structure containing cysteine-rich motifs. LTBP-2 is an integral component of elastin-containing microfibrils and contains 20 EGF-like repeats and four copies of an 8-cysteine repeat. LTBP-2 is synthesized as a protein by human foreskin fibroblasts. LTBP-2 co-localizes with tropoelastin in several tissues, including lung, dermis, epicardium, pericardium and heart valves, throughout rodent development, and in the spleen in the young adult mouse. Pseudoexfoliation (PEX) syndrome is a systemic condition characterized by the pathologic production and accumulation of an abnormal fibrillar extracellular material in many intra- and extraocular tissues. The co-localization of LTBP-1 and LTBP-2 with latent TGF β 1 and with fibrillin-1 on PEX fibrils suggests a possible mechanism for the regulation of TGF β 1 activity in PEX eyes. The LTBP-2 gene maps to human chromosome 14q24.3.

REFERENCES

- Moren, A., et al. 1994. Identification and characterization of LTBP-2, a novel latent transforming growth factor- β -binding protein. *J. Biol. Chem.* 269: 32469-32478.
- Bashir, M.M., et al. 1996. Analysis of the human gene encoding latent transforming growth factor- β -binding protein-2. *Int. J. Biochem. Cell Biol.* 28: 531-542.
- Shipley, J.M., et al. 2000. Developmental expression of latent transforming growth factor β binding protein 2 and its requirement early in mouse development. *Mol. Cell. Biol.* 20: 4879-4887.
- Schlotzer-Schrehardt, U., et al. 2001. Role of transforming growth factor- β 1 and its latent form binding protein in pseudoexfoliation syndrome. *Exp. Eye Res.* 73: 765-780.

CHROMOSOMAL LOCATION

Genetic locus: LTBP2 (human) mapping to 14q24.3.

PRODUCT

LTBP-2 shRNA (h) 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 μ l frozen stock containing 1.0×10^6 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 LTBP-2 siRNA (h): sc-43388 and LTBP-2 shRNA Plasmid (h): sc-43388-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.

PROTOCOLS

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

APPLICATIONS

LTBP-2 shRNA (h) Lentiviral Particles is recommended for the inhibition of LTBP-2 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 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

LTBP-2 (E-10): sc-166199 is recommended as a control antibody for monitoring of LTBP-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 LTBP-2 gene expression knockdown using RT-PCR Primer: LTBP-2 (h)-PR: sc-43388-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

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.