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LOXL1 shRNA (h) Lentiviral Particles: sc-45220-V

BACKGROUND

Lysyl oxidase (LOX) proteins belong to a family of enzymes that oxidize primary amine substrates to reactive aldehydes. In fibrillar collagens and elastin, LOX catalyzes the lysine-derived cross-links of collagen fibrils and insoluble elastic fibers in the extracellular matrix. It can localize both to the nucleus and the cytoplasm. LOX is involved in tumor suppression, cell motility, cellular senescence and developmental regulation. There are four homologs of LOX, lysyl oxidase-like proteins, designated LOX-like (LOXL1-LOXL4) proteins. LOXL1 is an extracellular protein that localizes specifically to sites of elastogenesis. It serves as a cross-linking enzyme, controlling the deposition of elastin. LOXL1 interacts with Fibulin-5.

REFERENCES

1. Kenyon, K., et al. 1993. A novel human cDNA with a predicted protein similar to lysyl oxidase maps to chromosome 15q24-q25. *J. Biol. Chem.* 268: 18435-18437.
2. Goy, A., et al. 2000. Physical linkage of the lysyl oxidase-like (LOXL1) gene to the PML gene on human chromosome 15q22. *Cytogenet. Cell Genet.* 88: 22-24.
3. Csiszar, K., et al. 2001. Lysyl oxidases: a novel multifunctional amine oxidase family. *Prog. Nucleic Acid Res. Mol. Biol.* 70: 1-32.
4. Noblesse, E., et al. 2004. Lysyl oxidase-like and lysyl oxidase are present in the dermis and epidermis of a skin equivalent and in human skin and are associated to elastic fibers. *J. Invest. Dermatol.* 122: 621-630.
5. Hayashi, K., et al. 2004. Progressive hair loss and myocardial degeneration in rough coat mice: reduced lysyl oxidase-like (LOXL) in the skin and heart. *J. Invest. Dermatol.* 123: 864-871.

CHROMOSOMAL LOCATION

Genetic locus: LOXL1 (human) mapping to 15q24.1.

PRODUCT

LOXL1 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 LOXL1 siRNA (h): sc-45220 and LOXL1 shRNA Plasmid (h): sc-45220-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

LOXL1 shRNA (h) Lentiviral Particles is recommended for the inhibition of LOXL1 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

LOXL1 (H-11): sc-166632 is recommended as a control antibody for monitoring of LOXL1 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 LOXL1 gene expression knockdown using RT-PCR Primer: LOXL1 (h)-PR: sc-45220-PR (20 μ l, 413 bp). 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.