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LOXL4 shRNA (h) Lentiviral Particles: sc-45226-V

BACKGROUND

Lysyl oxidase (LOX) proteins belong to a family of enzymes that oxidize primary amine substrated to reactive aldehydes. In fibrillar collagens and elastin, LOX catalyzes the lysine-derived cross-links of collagen fibrils and insoluble elastic fibers within 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. LOXL4 is an extracellular protein that is widely expressed. Highest expression levels have been detected in testis, pancreas, cartilage and skeletal muscle.

REFERENCES

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2. Asuncion, L., et al. 2001. A novel human lysyl oxidase-like gene (LOXL4) on chromosome 10q24 has an altered scavenger receptor cysteine-rich domain. *Matrix Biol.* 20: 487-491.
3. Maki, J.M., et al. 2001. Cloning and characterization of a fifth human LOX isoenzyme: the third member of the LOX-related subfamily with four scavenger receptor cysteine-rich domains. *Matrix Biol.* 20: 493-496.
4. Kirschmann, D.A., et al. 2002. A molecular role for LOX in breast cancer invasion. *Cancer Res.* 62: 4478-4483.
5. Bronson, N.W., et al. 2005. LOXL null mice demonstrate selective dentate structural changes but maintain dentate granule cell and CA1 pyramidal cell potentiation in the hippocampus. *Neurosci. Lett.* 390: 118-122.
6. Kim, D.J., et al. 2008. Lysyl oxidase like 4, a novel target gene of TGFβ1 signaling, can negatively regulate TGFβ1-induced cell motility in PLC/PRF/5 hepatoma cells. *Biochem. Biophys. Res. Commun.* 373: 521-527.

CHROMOSOMAL LOCATION

Genetic locus: LOXL4 (human) mapping to 10q24.2.

PRODUCT

LOXL4 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 μl frozen stock containing 1.0 x 10⁶ 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 LOXL4 siRNA (h): sc-45226 and LOXL4 shRNA Plasmid (h): sc-45226-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

LOXL4 shRNA (h) Lentiviral Particles is recommended for the inhibition of LOXL4 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μl frozen viral stock containing 1.0 x 10⁶ 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

LOXL4 (D-11): sc-374121 is recommended as a control antibody for monitoring of LOXL4 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 LOXL4 gene expression knockdown using RT-PCR Primer: LOXL4 (h)-PR: sc-45226-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.