

LOXL3 siRNA (h): sc-45224

BACKGROUND

Lysyl oxidase (LOX) proteins belong to a family of enzymes that oxidase 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 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. LOXL3 is an extracellular protein that localizes specifically to sites of elastogenesis. LOXL2 and LOXL3 can interact and cooperate with the SNAIL protein to down-regulate E-cadherin expression. In epithelial cells, overexpression of LOXL2 or LOXL3 may induce an epithelial-mesenchymal transitions process, an important element in tumor progression. LOXL3 is a widely expressed protein with highest levels of expression in placenta, small intestine, testis, heart, ovary and spleen.

REFERENCES

1. Jourdan-Le Saux, C., et al. 1999. The LOXL2 gene encodes a new LOX-like protein and is expressed at high levels in reproductive tissues. *J. Biol. Chem.* 274: 12939-12944.
2. Huang, Y., et al. 2001. Cloning and characterization of a human lysyl oxidase-like 3 gene (hLOXL3). *Matrix Biol.* 20: 153-157.
3. Jourdan-Le Saux, C., et al. 2001. Central nervous system, uterus, heart and leukocyte expression of the LOXL3 gene, encoding a novel LOX-like protein. *Genomics* 74: 211-218.
4. Maki, J.M., et al. 2001. Cloning and characterization of a fourth human lysyl oxidase isoenzyme. *Biochem. J.* 355: 381-387.
5. Molnar, J., et al. 2003. Structural and functional diversity of lysyl oxidase and the LOX-like proteins. *Biochim. Biophys. Acta* 1647: 220-224.
6. 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.
7. Peinado, H., et al. 2005. A molecular role for lysyl oxidase-like 2 enzyme in snail regulation and tumor progression. *EMBO J.* 24: 3446-3458.

CHROMOSOMAL LOCATION

Genetic locus: LOXL3 (human) mapping to 2p13.1.

PRODUCT

LOXL3 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see LOXL3 shRNA Plasmid (h): sc-45224-SH and LOXL3 shRNA (h) Lentiviral Particles: sc-45224-V as alternate gene silencing products.

For independent verification of LOXL3 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45224A, sc-45224B and sc-45224C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNases and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

LOXL3 siRNA (h) is recommended for the inhibition of LOXL3 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 μ M in 66 μ l. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

LOXL3 (E-6): sc-377216 is recommended as a control antibody for monitoring of LOXL3 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 reagents are recommended: 1) Western Blotting: use m-IgG κ BP-HRP: sc-516102 or m-IgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG κ BP-FITC: sc-516140 or m-IgG κ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor LOXL3 gene expression knockdown using RT-PCR Primer: LOXL3 (h)-PR: sc-45224-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

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