SANTA CRUZ BIOTECHNOLOGY, INC.

LLH2 siRNA (m): sc-60951

BACKGROUND

Lysyl hydroxylases (LLHs) 1-3 are hydroxylysines that function as attachment sites for carbohydrates. In collagen, the LLHs form hydroxylysine residues in -Xaa-Lys-Gly- sequences and are crucial for collagen crosslink stability. They form homodimers that localize to the endoplasmic reticulum. LLH1 is strongly expressed in liver, heart, lung, skeletal muscle and kidney tissue. LLH2 is highly expressed in heart, lung, kidney, eye, ovary and placenta, whereas LLH3 is expressed mainly in heart, lung, liver and testis. LLH2 exists in two alternatively-spliced forms: the long transcript (the major ubiquitously-expressed form) and the short transcript (expressed only in human kidney, spleen, liver and placenta). The long form of LLH2 is a bifunctional protein that plays a pivotal role in fibrosis by directing the collagen cross-link pattern. Any type of disruption of the regulation of the LLH2 transcript splicing may affect the stability of the extracellular matrix and contribute to specific connective tissue disorders.

REFERENCES

1. Yeowell, H.N. and Walker, L.C. 1999. Tissue specificity of a new splice form of the human lysyl hydroxylase 2 gene. Matrix Biol. 18: 179-187.
2. Ruotsalainen, H., et al. 1999. Characterization of cDNAs for mouse lysyl hydroxylase 1, 2 and 3, their phylogenetic analysis and tissue-specific expression in the mouse. Matrix Biol. 18: 325-329.
3. Ruotsalainen, H., et al. 2001. Complete genomic structure of mouse lysyl hydroxylase 2 and 3/collagen glucosyltransferase. Matrix Biol. 20: 137-146.
4. Risteli, M., et al. 2004. Characterization of collagenous peptides bound to lysyl hydroxylase isoforms. J. Biol. Chem. 279: 37535-37543.
5. Walker, L.C., et al. 2005. Tissue-specific expression and regulation of the alternatively-spliced forms of lysyl hydroxylase 2 (LH2) in human kidney cells and skin fibroblasts. Matrix Biol. 23: 515-523.
6. Zuurmond, A.M., et al. 2005. Minoxidil exerts different inhibitory effects on gene expression of lysyl hydroxylase 1, 2, and 3: implications for collagen cross-linking and treatment of fibrosis. Matrix Biol. 24: 261-270.
7. Brinckmann, J., et al. 2005. Interleukin 4 and prolonged hypoxia induce a higher gene expression of lysyl hydroxylase 2 and an altered cross-link pattern: important pathogenetic steps in early and late stage of systemic scleroderma? Matrix Biol. 24: 459-468.
8. Takashi, M., et al. 2005. Differential gene expression of collagen-binding small leucine-rich proteoglycans and lysyl hydroxylases, during mineralization by MC3T3-E1 cells cultured on titanium implant material. Eur. J. Oral Sci. 113: 225-231.
9. Wu, J., et al. 2006. Functional diversity of lysyl hydroxylase 2 in collagen synthesis of human dermal fibroblasts. Exp. Cell Res. 312: 3485-3494.

CHROMOSOMAL LOCATION

Genetic locus: Plod2 (mouse) mapping to 9 E3.3.

PROTOCOLS

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

PRODUCT

LLH2 siRNA (m) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see LLH2 shRNA Plasmid (m): sc-60951-SH and LLH2 shRNA (m) Lentiviral Particles: sc-60951-V as alternate gene silencing products.

For independent verification of LLH2 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60951A, sc-60951B and sc-60951C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCL, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

LLH2 siRNA (m) is recommended for the inhibition of LLH2 expression in mouse 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 µM in 66 µl. 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor LLH2 gene expression knockdown using RT-PCR Primer: LLH2 (m)-PR: sc-60951-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Liu, W., et al. 2018. Lysyl hydroxylases are transcription targets for GATA3 driving lung cancer cell metastasis. Sci. Rep. 8: 11905.

RESEARCH USE

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