

LAL siRNA (h): sc-72119

BACKGROUND

The Lipase gene family is part of one of the largest genetic superfamilies in living organisms. Members of the AB hydrolase subfamily all contain an enzyme core with an α/β sheet, not barrel, of eight β -sheets connected by α -helices. The AB hydrolase subfamily plays a crucial role in the metabolism of lipids. Members of this family include hepatic lipase (HL), endothelial lipase (EL), lipoprotein lipase (LPL), pancreatic lipase (PL), gastric lipase (GL), LCAT, and lysosomal acid lipase (LAL). LAL is the important enzyme in the hydrolysis of triglycerides and cholesteryl esters in lysosomes. LAL has six potential N-glycosylation sites and one potential O-glycosylation site, and it is mediated by macrophage mannose receptors. Defects in the LAL gene have been linked to Wolman disease (WD) and CE storage disease (CESD), while over-expression of LAL leads to atherosclerosis.

REFERENCES

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2. Du, H., et al. 1998. Targeted disruption of the mouse lysosomal acid lipase gene: long-term survival with massive cholesteryl ester and triglyceride storage. *Hum. Mol. Genet.* 7: 1347-1354.
3. Du, H., et al. 2001. Lysosomal acid lipase-deficient mice: depletion of white and brown fat, severe hepatosplenomegaly, and shortened life span. *J. Lipid Res.* 42: 489-500.
4. Du, H., et al. 2001. Enzyme therapy for lysosomal acid lipase deficiency in the mouse. *Hum. Mol. Genet.* 10: 1639-1648.
5. Du, H., et al. 2005. The role of mannosylated enzyme and the mannose receptor in enzyme replacement therapy. *Am. J. Hum. Genet.* 77: 1061-1074.
6. Ikeda, S., et al. 2005. Production of recombinant human lysosomal acid lipase in *Schizosaccharomyces pombe*: development of a fed-batch fermentation and purification process. *J. Biosci. Bioeng.* 98: 366-373.
7. Tadiboyina, V.T., et al. 2005. Treatment cholesterol ester storage disease. *Lipids Health Dis.* 4: 26.

CHROMOSOMAL LOCATION

Genetic locus: LIPA (human) mapping to 10q23.31.

PRODUCT

LAL 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 LAL shRNA Plasmid (h): sc-72119-SH and LAL shRNA (h) Lentiviral Particles: sc-72119-V as alternate gene silencing products.

For independent verification of LAL (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-72119A, sc-72119B and sc-72119C.

RESEARCH USE

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

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

LAL siRNA (h) is recommended for the inhibition of LAL 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

LAL (9G7F12): sc-58374 is recommended as a control antibody for monitoring of LAL 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 LAL gene expression knockdown using RT-PCR Primer: LAL (h)-PR: sc-72119-PR (20 μ l, 487 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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