SANTA CRUZ BIOTECHNOLOGY, INC.

LCAT siRNA (h): sc-60926

BACKGROUND

The lipase gene family belongs to one of the most robust genetic superfamilies found in living organisms, which includes esterases and thioesterases. Members of the AB hydrolase subfamily include hepatic lipase (HL), endothelial lipase (EL), lipoprotein lipase (LPL), pancreatic lipase (PL), gastric lipase (GL) and the lecithin-cholesterol acyltransferase (LCAT). These family members play a crucial role in the metabolism of lipids. LCAT esterifies cholesterol, which is required for cholesterol transport. LCAT deficiency has been implicated in fish-eye disease, a rare genetic disorder of high density lipoprotein (HDL) metabolism.

REFERENCES

1. McIntyre, N. 1988. Familial LCAT deficiency and fish-eye disease. J. Inherit. Metab. Dis. 1: 45-56.
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3. Huesca-Gomez, C., Carreon-Torres, E., Nepomuceno-Mejia, T., Sanchez-Solorio, M., Galicia-Hidalgo, M., Mejia, A.M., Montano, L.F., Franco, M., Posadas-Romero, C. and Perez-Mendez, O. 2004. Contribution of cholesteryl ester transfer protein and lecithin:cholesterol acyltransferase to HDL size distribution. Endocr. Res. 30: 403-415.
4. Nakamura, Y., Kotite, L., Gan, Y., Spencer, T.A., Fielding, C.J. and Fielding, P.E. 2004. Molecular mechanism of reverse cholesterol transport: reaction of pre-β-migrating high-density lipoprotein with plasma lecithin/cholesterol acyltransferase. Biochemistry 43: 14811-14820.
5. Miida, T., Zhang, B., Obayashi, K., Seino, U., Zhu, Y., Ito, T., Nakamura, Y., Okada, M. and Saku, K. 2004. T13M mutation of lecithin-cholesterol acyltransferase gene causes fish-eye disease. Clin. Chim. Acta 343: 201-208.

CHROMOSOMAL LOCATION

Genetic locus: LCAT (human) mapping to 16q22.1.

PRODUCT

LCAT 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see LCAT shRNA Plasmid (h): sc-60926-SH and LCAT shRNA (h) Lentiviral Particles: sc-60926-V as alternate gene silencing products.

For independent verification of LCAT (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-60926A, sc-60926B and sc-60926C.

PROTOCOLS

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

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

LCAT siRNA (h) is recommended for the inhibition of LCAT 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 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.

GENE EXPRESSION MONITORING

LCAT (D-2): sc-376682 is recommended as a control antibody for monitoring of LCAT 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 LCAT gene expression knockdown using RT-PCR Primer: LCAT (h)-PR: sc-60926-PR (20 µl). 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.