SANTA CRUZ BIOTECHNOLOGY, INC.

LTA4H siRNA (m): sc-42897

BACKGROUND

Leukotrienes are biologically active compounds formed from arachidonic acid or polyunsaturated fatty acids that are important in host defense reactions and play a pathophysiological role in inflammation and allergic reactions. LTA4H (leukotriene A_4-hydrolase) is a Zn^{2+}-containing enzyme with both epoxide hydrolase and aminopeptidase activity. As an epoxide hydrolase, LTA4H catalyzes the hydration of LTA4 to leukotriene B_4 (LTB4, 5S,12R-dihydroxy-6,14-*cis*-8,10-*trans*-eicosatetraenoic acid), a potent lipid chemoattractant that influences inflammation, immune responses and host defense against infection. As an aminopeptidase, LTA4H catalyzes the cleavage of amides of paranitroaniline. The human LTA4H gene encodes a 610 amino acid protein.

REFERENCES

1. Minami, M., et al. 1987. Molecular cloning of a cDNA coding for human leukotriene A_4 hydrolase. Complete primary structure of an enzyme involved in eicosanoid synthesis. J. Biol. Chem. 262: 13873-13876.
2. Funk, C.D., et al. 1987. Molecular cloning and amino acid sequence of leukotriene A4 hydrolase. Proc. Natl. Acad. Sci. USA 84: 6677-6681.
3. Gierse, J.K., et al. 1993. High-level expression and purification of human leukotriene A_4 hydrolase from insect cells infected with a baculovirus vector. Protein Expr. Purif. 4: 358-366.
4. Parnas, B.L., et al. 1996. Isolation and structure of leukotriene-A_4 hydrolase inhibitor: 8(S)-amino-2(R)-methyl-7-oxononanoic acid produced by *Streptomyces diastaticus*. J. Nat. Prod. 59: 962-964.
5. Online Mendelian Inheritance in Man, OMIM™. 1998. Johns Hopkins University, Baltimore, MD. MIM Number: 151570. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Thunnissen, M.M., et al. 2001. Crystal structure of human leukotriene A_4 hydrolase, a bifunctional enzyme in inflammation. Nat. Struct. Biol. 8: 131-135.
7. Rudberg, P.C., et al. 2002. Leukotriene A_4 hydrolase: selective abrogation of leukotriene B_4 formation by mutation of aspartic acid 375. Proc. Natl. Acad. Sci. USA 99: 4215-4220.
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CHROMOSOMAL LOCATION

Genetic locus: Lta4h (mouse) mapping to 10 C2.

PRODUCT

LTA4H 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 LTA4H shRNA Plasmid (m): sc-42897-SH and LTA4H shRNA (m) Lentiviral Particles: sc-42897-V as alternate gene silencing products.

For independent verification of LTA4H (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-42897A, sc-42897B and sc-42897C.

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

LTA4H siRNA (m) is recommended for the inhibition of LTA4H 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.

GENE EXPRESSION MONITORING

LTA4H (D-6): sc-390567 is recommended as a control antibody for monitoring of LTA4H 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 LTA4H gene expression knockdown using RT-PCR Primer: LTA4H (m)-PR: sc-42897-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.

PROTOCOLS

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