

5-LO siRNA (h): sc-29596

BACKGROUND

5-lipoxygenase (5-LO) is expressed primarily in polymorphonuclear leukocytes, macrophages, and mast cells. 5-LO performs the first two catalytic reactions in the biosynthesis of leukotrienes, lipid metabolites that induce contractions of airway smooth muscle and increase vascular permeability during anaphylaxis. The cellular localization of 5-LO varies between cell types. In activated blood polymorphonuclear leukocytes 5-LO undergoes calcium dependent translocation from the cytosol to the nuclear envelope. In alveolar macrophages, the majority of 5-LO is localized in the nucleus and, upon activation of these cells, intranuclear 5-LO binds to the nuclear membrane. This intracellular shuttling of 5-LO is dependent on the association with various signaling molecules, phosphorylation and the presence of a distinct nuclear localization signal, which is encoded at the amino terminus of 5-LO.

CHROMOSOMAL LOCATION

Genetic locus: ALOX5 (human) mapping to 10q11.21.

PRODUCT

5-LO 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 5-LO shRNA Plasmid (h): sc-29596-SH and 5-LO shRNA (h) Lentiviral Particles: sc-29596-V as alternate gene silencing products.

For independent verification of 5-LO (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-29596A, sc-29596B and sc-29596C.

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

5-LO siRNA (h) is recommended for the inhibition of 5-LO 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

5-LO (33): sc-136195 is recommended as a control antibody for monitoring of 5-LO 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 5-LO gene expression knockdown using RT-PCR Primer: 5-LO (h)-PR: sc-29596-PR (20 μ l, 542 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Sveinbjörnsson, B., et al. 2008. Expression of enzymes and receptors of the leukotriene pathway in human neuroblastoma promotes tumor survival and provides a target for therapy. *FASEB J.* 22: 3525-3536.
- Giannoni, E., et al. 2009. Redox regulation of anoikis resistance of metastatic prostate cancer cells: key role for Src and EGFR-mediated pro-survival signals. *Oncogene* 28: 2074-2086.
- Lim, J.Y., et al. 2010. MK886-induced apoptosis depends on the 5-LO expression level in human malignant glioma cells. *J. Neurooncol.* 97: 339-346.
- Tang, F., et al. 2015. Cysteinyl leukotrienes mediate lymphokine killer activity induced by NKG2D and IL-15 in cytotoxic T cells during celiac disease. *J. Exp. Med.* 212: 1487-1495.
- Woo, S.M., et al. 2017. Up-regulation of 5-lipoxygenase by inhibition of cathepsin G enhances TRAIL-induced apoptosis through down-regulation of survivin. *Oncotarget* 8: 106672-106684.
- Park, M., et al. 2022. Zileuton alleviates radiation-induced cutaneous ulcers via inhibition of senescence-associated secretory phenotype in rodents. *Int. J. Mol. Sci.* 23: 8390.

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.