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CysLT₂ Receptor siRNA (h): sc-43713

BACKGROUND

Cysteinyl leukotriene (CysLTs) induce intracellular calcium mobilization through the binding of two distinct seven-transmembrane, G protein-coupled receptors, designated CysLT₁ and CysLT₂ Receptors, to induce potent bronchoconstriction. Airway smooth muscle and macrophages express both receptor types, and additionally monocytes and eosinophils express CysLT₁ Receptor, while cardiac Purkinje cells, adrenal medulla, peripheral blood leukocytes and brain also utilize CysLT₂ Receptor. The effects of the CysLT receptors can be blocked by antagonists, indicating a therapeutic mechanism for the treatment of asthma and allergies.

REFERENCES

1. Sarau, H.M., et al. 1999. Identification, molecular cloning, expression, and characterization of a cysteinyl leukotriene receptor. *Mol. Pharmacol.* 56: 657-663.
2. Lynch, K.R., et al. 1999. Characterization of the human cysteinyl leukotriene CysLT₁ Receptor. *Nature* 399: 789-793.
3. Heise, C.E., et al. 2000. Characterization of the human cysteinyl leukotriene 2 receptor. *J. Biol. Chem.* 275: 30531-30536.
4. Sjostrom, M., et al. 2001. Human umbilical vein endothelial cells generate leukotriene C4 via microsomal glutathione S-transferase type 2 and express the CysLT₁ Receptor. *Eur. J. Biochem.* 268: 2578-2586.
5. Maekawa, A., et al. 2001. Identification in mice of two isoforms of the cysteinyl leukotriene 1 receptor that result from alternative splicing. *Proc. Natl. Acad. Sci. USA* 98: 2256-2261.
6. Leff, A.R. 2001. Regulation of leukotrienes in the management of asthma: biology and clinical therapy. *Annu. Rev. Med.* 52: 1-14.

CHROMOSOMAL LOCATION

Genetic locus: CYSLTR2 (human) mapping to 13q14.2.

PRODUCT

CysLT₂ Receptor 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 CysLT₂ Receptor shRNA Plasmid (h): sc-43713-SH and CysLT₂ Receptor shRNA (h) Lentiviral Particles: sc-43713-V as alternate gene silencing products.

For independent verification of CysLT₂ Receptor (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-43713A, sc-43713B and sc-43713C.

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.

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

CysLT₂ Receptor siRNA (h) is recommended for the inhibition of CysLT₂ Receptor 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

CysLT₂ Receptor (B-7): sc-514181 is recommended as a control antibody for monitoring of CysLT₂ Receptor 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 CysLT₂ Receptor gene expression knockdown using RT-PCR Primer: CysLT₂ Receptor (h)-PR: sc-43713-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.