SANTA CRUZ BIOTECHNOLOGY, INC.

IL-5 siRNA (m): sc-39626

BACKGROUND

Interleukin-5, or IL-5, was originally discovered as a soluble T cell-derived factor, called T cell-replacing factor (TRF), that induced T cell-depleted activated B cells to secrete immunoglobulin. Native IL-5 is a disulfide-linked homodimer. IL-5 is initially synthesized as a precursor with a 19 amino acid signal peptide which is cleaved to form a 112 amino acid mature protein. Murine and human IL-5 exhibit 70% sequence identity at the amino acid level. IL-5 exerts its biological activity through the IL-5 receptor (IL-5R), which is composed of at least two chains: an α chain that binds IL-5 with low affinity and a β chain that does not bind IL-5, but together with the IL-5 α chain, constitutes the high affinity IL-5 receptor. The β chain is common to the IL-3, IL-5 and GM-CSF receptors and has been shown to signal through the JAK/Stat pathway.

REFERENCES

1. Takatsu, K., et al. 1980. Antigen-induced T cell-replacing factor (TRF). I. Functional characterization of a TRF-producing helper T cell subset and genetic studies on TRF production. J. Immunol. 124: 2414-2422.
2. Azuma, C., et al. 1986. Cloning of cDNA for human T cell replacing factor (interleukin-5) and comparison with the murine homologue. Nucleic Acids Res. 14: 9149-9158.
3. Li, J., et al. 1996. Single chain human interleukin 5 and its asymmetric mutagenesis for mapping receptor binding sites. J. Biol. Chem. 271: 1817-1820.
4. Freeburn, R.W., et al. 1996. The β subunit common to the GM-CSF, IL-3 and IL-5 receptors is highly polymorphic but pathogenic point mutations in patients with acute myeloid leukaemia (AML) are rare. Leukemia 10: 123-129.
5. Sun, Z., et al. 1996. Interleukin-5 receptor α subunit gene regulation in human eosinophil development: identification of a unique ***cis***-element that acts like an enhancer in regulating activity of the IL-5Rα promoter. Curr. Top. Micro. Immunol. 211: 173-187.
6. Bates, M.E., et al. 1996. IL-5 activates a 45-kDa mitogen-activated protein (MAP) kinase and JAK2 tyrosine kinase in human eosinophils. J. Immunol. 156: 711-718.

CHROMOSOMAL LOCATION

Genetic locus: Il5 (mouse) mapping to 11 B1.3.

PRODUCT

IL-5 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 IL-5 shRNA Plasmid (m): sc-39626-SH and IL-5 shRNA (m) Lentiviral Particles: sc-39626-V as alternate gene silencing products.

For independent verification of IL-5 (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-39626A, sc-39626B and sc-39626C.

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

IL-5 siRNA (m) is recommended for the inhibition of IL-5 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

IL-5 (B-2): sc-398334 is recommended as a control antibody for monitoring of IL-5 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 IL-5 gene expression knockdown using RT-PCR Primer: IL-5 (m)-PR: sc-39626-PR (20 µl, 502 bp). 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.