SANTA CRUZ BIOTECHNOLOGY, INC.

IL-3 siRNA (m): sc-39622

BACKGROUND

Interleukin-3, or IL-3, is a pleiotropic cytokine that is primarily secreted by activated T lymphocytes and stimulates the proliferation and differentiation of hematopoietic cells. IL-3 not only supports growth of both pluripotent stem cells and the more differentiated committed progenitors, but it also stimulates the functional activity of some fully differentiated cells. IL-3 has also been shown to protect mast cells from undergoing apoptosis. IL-3 exerts its biological effects through a receptor which consists of a ligand-specific α subunit and a signal transducing β subunit common to the IL-3/IL-5/GM-CSF receptors. The carboxy-terminus of the β subunit has been shown to be necessary for activation of the MAP kinase signaling pathway. Although the IL-3 receptor has no intrinsic kinase activity, stimulation with IL-3 leads to tyrosine phosphorylation of the JAK/Tyk 2 family member, JAK2, which in turn activates and causes nuclear translocation of Stat5a and Stat5b.

REFERENCES

1. Abrams, J.S. and Pearce, M.K. 1988. Development of rat anti-mouse interleukin 3 monoclonal antibodies which neutralize bioactivity *in vitro*. J. Immunol. 140: 131-137.
2. Cockayne, D.A., et al. 1992. Antisense RNA inhibition of hematopoietic growth factor production. Growth Factors 5: 171-181.
3. Abrams, J.S., et al. 1992. Strategies of anti-cytokine monoclonal antibody development: immunoassay of IL-10 and IL-5 in clinical samples. Immunol. Rev. 127: 5-24.
4. Magnelli, L., et al. 1993. Apoptosis induction in 32D cells by IL-3 withdrawal is preceded by a drop in the intracellular calcium level. Biochem. Biophys. Res. Commun. 194: 1394-1397.
5. Sander, B., et al. 1994. Similar frequencies and kinetics of cytokine producing cells in murine peripheral blood and spleen. Cytokine detection by immunoassay and intracellular immunostaining. J. Immunol. Methods 166: 201-214.
6. Kinoshita, T., et al. 1995. Suppression of apoptotic death in hematopoietic cells by signalling through the IL-3/GM-CSF receptors. EMBO J. 14: 266-275.

CHROMOSOMAL LOCATION

Genetic locus: Il3 (mouse) mapping to 11 B1.3.

PRODUCT

IL-3 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-3 shRNA Plasmid (m): sc-39622-SH and IL-3 shRNA (m) Lentiviral Particles: sc-39622-V as alternate gene silencing products.

For independent verification of IL-3 (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-39622A, sc-39622B and sc-39622C.

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-3 siRNA (m) is recommended for the inhibition of IL-3 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-3 (G-11): sc-398210 is recommended as a control antibody for monitoring of IL-3 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-3 gene expression knockdown using RT-PCR Primer: IL-3 (m)-PR: sc-39622-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.