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IL-6 siRNA (r): sc-156148

BACKGROUND

Interleukin-6, or IL-6, is a multifunctional protein, 212 amino acids in length, that plays critical roles in host defense, immune response and hematopoiesis. IL-6 is constitutively expressed by epidermal Langerhans cells and its expression is induced in stimulated keratinocytes. IL-6, IL-1 β and TNF α act as endogenous pyrogens, regulating the fever response to bacterial invasion. The IL-6 receptor is a trimeric complex composed of an IL-6-specific α chain and a homodimer of the gp130 glycoprotein common to the IL-6, IL-11, CNTF, OSM and LIF receptors. Stimulation with IL-6 leads to gp130 homodimerization and the activation of associated kinases JAK1 and JAK2. Once activated, JAK1 and JAK2 phosphorylate Stat3, causing its nuclear translocation and transcription of Stat3-responsive genes. IL-6 has also been shown to activate the Ras/MAP kinase pathway, which regulates NFIL6 transcription.

REFERENCES

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- Zhong, Z., et al. 1994. Stat3: A Stat family member activated by tyrosine phosphorylation in response to epidermal growth factor and interleukin-6. *Science* 264: 95-98.
- Wijdenes, J., et al. 1995. Interleukin-6 signal transducer gp130 has specific binding sites for different cytokines as determined by antagonistic and agonistic anti-gp130 monoclonal antibodies. *Eur. J. Immunol.* 25: 3474-3481.
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- Holliday, M.R., et al. 1996. Stimulation by oxazolone of increased IL-6, but not IL-10, in the skin of mice. *Toxicology* 106: 237-242.
- Chai, Z., et al. 1996. Interleukin (IL)-6 gene expression in the central nervous system is necessary for fever response to lipopolysaccharide or IL-1 β : a study on IL-6-deficient mice. *J. Exp. Med.* 183: 311-316.

CHROMOSOMAL LOCATION

Genetic locus: Il6 (rat) mapping to 4q11.

PRODUCT

IL-6 siRNA (r) 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 IL-6 shRNA Plasmid (r): sc-156148-SH and IL-6 shRNA (r) Lentiviral Particles: sc-156148-V as alternate gene silencing products.

For independent verification of IL-6 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-156148A, sc-156148B and sc-156148C.

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

IL-6 siRNA (r) is recommended for the inhibition of IL-6 expression in rat 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

IL-6 (10E5): sc-57315 is recommended as a control antibody for monitoring of IL-6 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 IL-6 gene expression knockdown using RT-PCR Primer: IL-6 (r)-PR: sc-156148-PR (20 μ l, 546 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.