

LIFR siRNA (r): sc-270619

BACKGROUND

IL-6 activates intracellular signaling through binding a receptor consisting of a ligand-binding protein (IL-6R) and a second protein. IL-6 first binds to IL-6R which subsequently associates with a gp130 dimer. The active signaling complex consists of at minimum IL-6, IL-6R and a dimer of two gp130 proteins that are linked by a disulfide bond. A soluble form of IL-6R is generated by proteolytic cleavage of the membrane-bound precursor and can function as an agonistic molecule that can actively participate in cell-to-cell signaling. The second subunit of the IL-6 complex, gp130, also functions as a component of several additional receptor complexes including leukemia inhibitory factor (LIF), oncostatin M (OSM), ciliary neurotrophic factor (CNTF) and IL-11. LIF binds to the LIF receptor (LIFR) with low affinity and to a complex of the LIF receptor and gp130 with high affinity while OSM appears to bind to gp130 with low affinity and to a complex of gp130 and the LIF receptor with high affinity.

REFERENCES

1. Yamasaki, K., et al. 1988. Cloning and expression of the human interleukin-6 (BSF-2/IFN β 2) receptor. *Science* 241: 825-828.
2. Taga, T., et al. 1989. Interleukin-6 triggers the association of its receptor with a possible signal transducer, gp130. *Cell* 58: 573-581.
3. Hibi, M., et al. 1990. Molecular cloning and expression of an IL-6 signal transducer, gp130. *Cell* 63: 1149-1157.
4. Davis, S., et al. 1993. LIFR β and gp130 as heterodimerizing signal transducers of the tripartite CNTF receptor. *Science* 260: 1805-1808.
5. Murakami, M., et al. 1993. Critical cytoplasmic region of the interleukin-6 signal transducer gp130 is conserved in the cytokine receptor family. *Science* 260: 1808-1810.
6. Müllberg, J., et al. 1994. The soluble human IL-6 receptor. Mutational characterization of the proteolytic cleavage site. *J. Immunol.* 152: 4958-4968.
7. Kishimoto, T., et al. 1994. Cytokine signal transduction. *Cell* 76: 253-262.
8. Hilton, D.J., et al. 1994. Cloning of a murine IL-11 receptor α -chain; requirement for gp130 for high affinity binding and signal transduction. *EMBO J.* 13: 4765-4775.

CHROMOSOMAL LOCATION

Genetic locus: Lifr (rat) mapping to 2q16.

PRODUCT

LIFR 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 LIFR shRNA Plasmid (r): sc-270619-SH and LIFR shRNA (r) Lentiviral Particles: sc-270619-V as alternate gene silencing products.

For independent verification of LIFR (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-270619A, sc-270619B and sc-270619C.

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

LIFR siRNA (r) is recommended for the inhibition of LIFR 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

LIFR (A-10): sc-515337 is recommended as a control antibody for monitoring of LIFR 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 LIFR gene expression knockdown using RT-PCR Primer: LIFR (r)-PR: sc-270619-PR (20 μ l). 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.