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Stat3 shRNA (h2) Lentiviral Particles: sc-44275-V

BACKGROUND

Membrane receptor signaling by various ligands, including interferons and growth hormones such as EGF, induces activation of JAK kinases, which then leads to tyrosine phosphorylation of the various Stat transcription factors. Stat1 and Stat2 are induced by IFN- α and form a heterodimer, which is part of the ISGF-3 transcription factor complex. Although early reports indicate Stat3 activation by EGF and IL-6, it has been shown that Stat3 β appears to be activated by both while Stat3 α is activated by EGF, but not by IL-6. Highest expression of Stat4 is seen in testis and myeloid cells. IL-12 has been identified as an activator of Stat4. Stat5 has been shown to be activated by Prolactin and by IL-3. Stat6 is involved in IL-4 activated signaling pathways.

REFERENCES

1. 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.
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3. Hou, J., et al. 1994. An interleukin-4-induced transcription factor: IL-4 Stat. *Science* 265: 1701-1706.
4. Yamamoto, K., et al. 1994. Stat4, a novel γ interferon activation site-binding protein expressed in early myeloid differentiation. *Mol. Cell. Biol.* 14: 4342-4349.
5. Pallard, C., et al. 1995. Interleukin-3, erythropoietin, and Prolactin activate a Stat5-like factor in lymphoid cells. *J. Biol. Chem.* 270: 15942-15945.
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7. Schindler, C. et al. 1995. transcriptional responses to polypeptide ligands: the JAK-Stat pathway. *Annu. Rev. Biochem.* 64: 621-651.

CHROMOSOMAL LOCATION

Genetic locus: STAT3 (human) mapping to 17q21.2.

PRODUCT

Stat3 shRNA (h2) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Stat3 siRNA (h2): sc-44275 and Stat3 shRNA Plasmid (h2): sc-44275-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

Stat3 shRNA (h2) Lentiviral Particles is recommended for the inhibition of Stat3 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 lentiviral transducing particles per milliliter; contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Stat3 (C-20): sc-482 is recommended as a control antibody for monitoring of Stat3 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Stat3 gene expression knockdown using RT-PCR Primer: Stat3 (h2)-PR: sc-44275-PR (20 μ l, 500 bp). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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