

Stat5a siRNA (h): sc-37008

BACKGROUND

Signal transducer and activator of transcription 5A (Stat5a) and Stat5b, which share 96% homology, undergo receptor tyrosine kinase or G protein-coupled receptor-dependent phosphorylation in response to cytokines or growth factors, and then form homo- or heterodimers that translocate to the nucleus, where they initiate transcription. Activation of Stat5a via IL-2, IL-3, IL-7/ GM-CSF, erythropoietin, thrombopoietin and growth hormones influences proliferation, differentiation and apoptosis in lymphohematopoietic cells. Phosphorylation of Stat5a at Ser127/Ser128 and Ser779 are contingent on ErbB-4-mediated activation of Stat5a. Activation of Stat5b via IL-2, IL-4, CSF-1 and growth hormones influences TCR signaling, apoptosis, adult mammary gland development and sexual dimorphism of liver gene expression. Stat5b is the major liver-expressed Stat5 form that has been shown to fuse with the retinoic acid receptor a gene in acute promyelocytic leukemias (APLL). Stat5a/b null mice have severely impaired lymphoid development and differentiation.

CHROMOSOMAL LOCATION

Genetic locus: STAT5A (human) mapping to 17q21.2.

PRODUCT

Stat5a siRNA (h) 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 Stat5a shRNA Plasmid (h): sc-37008-SH and Stat5a shRNA (h) Lentiviral Particles: sc-37008-V as alternate gene silencing products.

For independent verification of Stat5a (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-37008A, sc-37008B and sc-37008C.

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

Stat5a siRNA (h) is recommended for the inhibition of Stat5a expression in human cells.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

Stat5a (C-6): sc-271542 is recommended as a control antibody for monitoring of Stat5a 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 Stat5a gene expression knockdown using RT-PCR Primer: Stat5a (h)-PR: sc-37008-PR (20 μ l, 598 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Lee, J.E., et al. 2012. Nongenomic Stat5-dependent effects on Golgi apparatus and endoplasmic reticulum structure and function. *Am. J. Physiol., Cell Physiol.* 302: C804-C820.
2. Lee, J.E., et al. 2013. Definitive evidence using enucleated cytoplasts for a nongenomic basis for the cystic change in endoplasmic reticulum structure caused by STAT5a/b siRNAs. *Am. J. Physiol., Cell Physiol.* 304: C312-C323.
3. Yang, Y.M., et al. 2013. Subcellular mechanisms in pulmonary arterial hypertension: combinatorial modalities that inhibit anterograde trafficking and cause bone morphogenetic protein receptor type 2 mislocalization. *Pulm. Circ.* 3: 533-550.
4. Chen, X.C., et al. 2017. EGF stimulates glioblastoma metastasis by induction of matrix metalloproteinase-9 in an EGFR-dependent mechanism. *Oncotarget* 8: 65969-65982.

RESEARCH USE

For research use only, not for use in diagnostic procedures.