

PIAS 3 siRNA (m): sc-37006

BACKGROUND

The IL-6-type family of cytokines, which includes IL-6 as well as a number of similar cytokines and growth factors, plays a significant role in regulating gene activation, proliferation and differentiation. Transcription factors of the STAT family are known to be involved in this signal transduction pathway, undergoing phosphorylation, dimerization, and translocation to the nucleus upon activation. PIAS 1, for protein inhibitor of activated Stat1 (also designated Gu/RNA helicase II binding protein), binds specifically to Stat1, blocking Stat1 DNA-binding activity and inhibiting Stat1-mediated gene activation. PIAS 1 also binds to the Gu/RNA helicase II enzyme, leading to the proteolytic cleavage of Gu/RH-II. PIAS 3 similarly binds specifically to Stat3, blocking Stat3 DNA-binding activity and inhibiting Stat3-mediated gene activation.

REFERENCES

1. Akira, S., et al. 1994. Molecular cloning of APRF, a novel IFN-stimulated gene factor 3 p91-related transcription factor involved in the gp130-mediated signaling pathway. *Cell* 77: 63-71.
2. 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.
3. Valdez, B.C., et al. 1997. Cloning and characterization of Gu/RH-II binding protein. *Biochem. Biophys. Res. Commun.* 234: 335-340.
4. Chung, C.D., et al. 1997. Specific inhibition of Stat3 signal transduction by PIAS 3. *Science* 278: 1803-1805.
5. Heinrich, P.C., et al. 1998. Interleukin-6-type cytokine signalling through the gp130/Jak/STAT pathway. *Biochem. J.* 334: 297-314.

CHROMOSOMAL LOCATION

Genetic locus: Pias3 (mouse) mapping to 3 F2.1.

PRODUCT

PIAS 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PIAS 3 shRNA Plasmid (m): sc-37006-SH and PIAS 3 shRNA (m) Lentiviral Particles: sc-37006-V as alternate gene silencing products.

For independent verification of PIAS 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-37006A, sc-37006B and sc-37006C.

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

PIAS 3 siRNA (m) is recommended for the inhibition of PIAS 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 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

PIAS 3 (C-12): sc-46682 is recommended as a control antibody for monitoring of PIAS 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-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 PIAS 3 gene expression knockdown using RT-PCR Primer: PIAS 3 (m)-PR: sc-37006-PR (20 μ l, 568 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Yoon, J.H., et al. 2015. Phosphorylation status determines the opposing functions of Smad2/Smad3 as STAT3 cofactors in TH17 differentiation. *Nat. Commun.* 6: 7600.

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.