

3BP2 siRNA (m): sc-40290

BACKGROUND

3BP2 is a Syk family kinase-interacting protein (SKIP) that is expressed in spleen and peripheral blood leukocytes. 3BP2 was originally characterized as an Abl SH3-interacting protein, as it contains a single proline-rich domain and an SH2 domain, consistent with other adaptor molecules. In Jurkat T cells transfected with 3BP2, stimulation of T-cell receptors (TCR) rapidly induces the redistribution of 3BP2 from the cytoplasm to the membrane, where it associates with the TCR/protein tyrosine kinase complexes. Through this translocation, 3BP2 is able to selectively bind to FIt3/FIk2 receptors and to the phosphorylated Syk, Lat and ZAP-70 proteins. In T lymphocytes, the overexpression of 3BP2, specifically the overexpression of the SH2 and proline rich domains, is sufficient to induce the activation of several transcription factors, including NFAT and AP-1. This transactivation results in the upregulation of the IL-2 gene promoter and suggests a role for 3BP2 in mediating T cell signaling.

REFERENCES

1. Ren, R., et al. 1993. Identification of a ten-amino acid proline-rich SH3 binding site. *Science* 259: 1157-1161.
2. Songyang, Z., et al. 1994. Specific motifs recognized by the SH2 domains of Csk, 3BP2, fps/fes, GRB-2, HCP, SHC, Syk, and Vav. *Mol. Cell. Biol.* 14: 2777-2785.
3. Bell, S.M., et al. 1997. Identification and characterization of the human homologue of SH3BP2, an SH3 binding domain protein within a common region of deletion at 4p16.3 involved in bladder cancer. *Genomics* 44: 163-170.
4. Deckert, M., et al. 1998. Adaptor function for the Syk kinases-interacting protein 3BP2 in IL-2 gene activation. *Immunity* 9: 595-605.
5. Shapiro, V.S., et al. 1998. Nuclear factor of activated T cells and AP-1 are insufficient for IL-2 promoter activation: requirement for CD28 up-regulation of RE/AP. *J. Immunol.* 161: 6455-6458.

CHROMOSOMAL LOCATION

Genetic locus: Sh3bp2 (mouse) mapping to 5 B2.

PRODUCT

3BP2 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 3BP2 shRNA Plasmid (m): sc-40290-SH and 3BP2 shRNA (m) Lentiviral Particles: sc-40290-V as alternate gene silencing products.

For independent verification of 3BP2 (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-40290A, sc-40290B and sc-40290C.

PROTOCOLS

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

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

3BP2 siRNA (m) is recommended for the inhibition of 3BP2 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

3BP2 (C-5): sc-166459 is recommended as a control antibody for monitoring of 3BP2 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 3BP2 gene expression knockdown using RT-PCR Primer: 3BP2 (m)-PR: sc-40290-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.