SANTA CRUZ BIOTECHNOLOGY, INC.

SH2-B siRNA (m): sc-40333

BACKGROUND

SH2-B is a component of the signaling network involved in the regulation of cell shape and movement. SH2-B is related to the APS (adapter molecule containing PH and SH2 domains) family of adapter proteins, which characteristically contain a Pleckstrin homology (PH) domain, an SH2 domain and a tyrosine phosphorylation site. SH2-B is alternatively spliced to generate three distinct isoforms, SH2-B α, β, and γ, that share an identical N-terminal sequence, including the PH domain, the SH2 domain and multiple proline-rich motifs. The isoform SH2-B β contributes to the regulation of the Actin cytoskeleton as it associates with various tyrosine kinases in response to growth factor stimulation. Following PDGF stimulation, SH2-B β can directly interact with the PDGF receptor (PDGFR), where it is phosphorylated on tyrosine residues and functions as a signaling protein for the PDGFR pathway. In addition, SH2-B β is also a substrate for JAK2 and, thereby, mediates the cytoskeletal reorganization that is induced by the signaling pathways of various growth factors.

REFERENCES

1. Frank, S.J., et al. 1995. Regions of the JAK2 tyrosine kinase required for coupling to the growth hormone receptor. J. Biol. Chem. 270: 14776-14785.
2. Rui, L., et al. 1997. Identification of SH2-B β as a substrate of the tyrosine kinase JAK2 involved in growth hormone signaling. Mol. Cell. Biol. 17: 6633-6644.
3. Rui, L. and Carter-Su, C. 1998. Platelet-derived growth factor (PDGF) stimulates the association of SH2-B β with PDGF receptor and phosphorylation of SH2-Bβ. J. Biol. Chem. 273: 21239-21245.
4. Rui, L. and Carter-Su, C. 1999. Identification of SH2-B β as a potent cytoplasmic activator of the tyrosine kinase Janus kinase 2. Proc. Natl. Acad. Sci. USA 96: 7172-7177.
5. Rui, L., et al 1999. SH2-B is required for nerve growth factor-induced neuronal differentiation. J. Biol. Chem. 274: 10590-10594.
6. Rui, L., et al. 1999. SH2-B, a membrane-associated adapter, is phosphorylated on multiple serines/threonines in response to nerve growth factor by kinases within the MEK/ERK cascade. J. Biol. Chem. 274: 26485-26492.

CHROMOSOMAL LOCATION

Genetic locus: Sh2b1 (mouse) mapping to 7 F3.

PRODUCT

SH2-B 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see SH2-B shRNA Plasmid (m): sc-40333-SH and SH2-B shRNA (m) Lentiviral Particles: sc-40333-V as alternate gene silencing products.

For independent verification of SH2-B (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-40333A, sc-40333B and sc-40333C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

SH2-B siRNA (m) is recommended for the inhibition of SH2-B 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 µM in 66 µl. 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

SH2-B $\alpha/\beta/\gamma/\delta$ (C-11): sc-514142 is recommended as a control antibody for monitoring of SH2-B 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 SH2-B gene expression knockdown using RT-PCR Primer: SH2-B (m)-PR: sc-40333-PR (20 µl). 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.