

PP2B-B2 siRNA (h): sc-92568

BACKGROUND

In eukaryotes, the phosphorylation and dephosphorylation of proteins on serine and threonine residues is an essential means of regulating a broad range of cellular functions, including division, homeostasis and apoptosis. A group of proteins that are intimately involved in this process are the protein phosphatases. In general, the protein phosphatase (PP) holoenzyme is a trimeric complex composed of a regulatory subunit, a variable subunit and a catalytic subunit. Four major families of protein phosphatase catalytic subunits have been identified, designated PP1, PP2A, PP2B and PP2C. An additional protein phosphatase catalytic subunit, PPX (also known as PP4), is a putative member of a novel PP family. The PP2B family comprises subfamily members PP2B-A α , PP2B-A β and PP2B-A γ . Two additional regulatory subunits have been identified, designated PP2B-B1 and PP2B-B2. PP2B-B2, also known as PPP3R2 (protein phosphatase 3, regulatory subunit B, β), PPP3RL, CBLP (calcineurin B-like protein) or CNBII (calcineurin BII), is a 170 amino acid protein that belongs to the calcineurin regulatory subunit family. Encoded by a gene that maps to human chromosome 9q31.1, PP2B-B2 is highly conserved. PP2B-B2 contains four EF-hand domains and exhibits testis-specific localization. PP2B-B2 is composed of a catalytic subunit (A) and a regulatory subunit (B), and functions as a regulatory subunit of calcineurin, a calcium-dependent, calmodulin stimulated protein phosphatase.

REFERENCES

1. Ueki, K., et al. 1992. Structure and expression of two isoforms of the murine calmodulin-dependent protein phosphatase regulatory subunit (calcineurin B). *Biochem. Biophys. Res. Commun.* 187: 537-543.
2. Cohen, P.T. 1993. Important roles for novel protein phosphatases dephosphorylating serine and threonine residues. *Biochem. Soc. Trans.* 21: 884-888.
3. Hendrix, P., et al. 1993. Structure and expression of a 72-kDa regulatory subunit of protein phosphatase 2A. Evidence for different size forms produced by alternative splicing. *J. Biol. Chem.* 268: 15267-15276.
4. Mumby, M.C. and Walter, G. 1993. Protein serine/threonine phosphatases: structure, regulation, and functions in cell growth. *Physiol. Rev.* 73: 673-699.
5. Okubo, S., et al. 1994. A regulatory subunit of smooth muscle myosin bound phosphatase. *Biochem. Biophys. Res. Commun.* 200: 429-434.

CHROMOSOMAL LOCATION

Genetic locus: PPP3R2 (human) mapping to 9q31.1.

PRODUCT

PP2B-B2 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 PP2B-B2 shRNA Plasmid (h): sc-92568-SH and PP2B-B2 shRNA (h) Lentiviral Particles: sc-92568-V as alternate gene silencing products.

For independent verification of PP2B-B2 (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-92568A, sc-92568B and sc-92568C.

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

PP2B-B2 siRNA (h) is recommended for the inhibition of PP2B-B2 expression in human 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

PP2B-B2 (MH-94): sc-134423 is recommended as a control antibody for monitoring of PP2B-B2 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 PP2B-B2 gene expression knockdown using RT-PCR Primer: PP2B-B2 (h)-PR: sc-92568-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.

PROTOCOLS

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