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PP2A-C α siRNA (r): sc-270193

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 (calcineurin) and PP2C. Characteristic of the protein phosphatase complexes, the PP2A phosphatase core enzyme is composed of a regulatory subunit and a catalytic subunit, the latter of which exists as two isoforms, designated PP2A α and PP2A β . The multiple subunits of PP2A work in concert to regulate a variety of metabolic pathways, including transcription, translation, cell cycle progression and oncogenic transformation.

REFERENCES

1. Strack, S., et al. 2002. Protein phosphatase 2A holoenzyme assembly: identification of contacts between B-family regulatory and scaffolding A subunits. *J. Biol. Chem.* 277: 20750-20755.
2. Avdi, N.J., et al. 2002. A role for protein phosphatase-2A in p38 mitogen-activated protein kinase-mediated regulation of the c-Jun NH₂-terminal kinase pathway in human neutrophils. *J. Biol. Chem.* 277: 40687-40696.
3. Zhou, J., et al. 2003. Characterization of the A α and A β subunit isoforms of protein phosphatase 2A: differences in expression, subunit interaction, and evolution. *Biochem. J.* 369: 387-398.
4. Shern, J.F., et al. 2003. Cytosolic Arl2 is complexed with cofactor D and protein phosphatase 2A. *J. Biol. Chem.* 278: 40829-40836.
5. Prickett, T.D., et al. 2004. Overlapping binding sites in protein phosphatase 2A for association with regulatory A and α -4 (mTap42) subunits. *J. Biol. Chem.* 279: 38912-38920.
6. Gergs, U., et al. 2004. Overexpression of the catalytic subunit of protein phosphatase 2A impairs cardiac function. *J. Biol. Chem.* 279: 40827-40834.

CHROMOSOMAL LOCATION

Genetic locus: Ppp2ca (rat) mapping to 10q22.

PRODUCT

PP2A-C α siRNA (r) is a pool of 2 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 PP2A-C α shRNA Plasmid (r): sc-270193-SH and PP2A-C α shRNA (r) Lentiviral Particles: sc-270193-V as alternate gene silencing products.

For independent verification of PP2A-C α (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270193A and sc-270193B.

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

PP2A-C α siRNA (r) is recommended for the inhibition of PP2A-C α expression in rat 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

PP2A-C α / β (1D6): sc-80665 is recommended as a control antibody for monitoring of PP2A-C α 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 MarkerTM 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 PP2A-C α gene expression knockdown using RT-PCR Primer: PP2A-C α (r)-PR: sc-270193-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.