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Diagnostik & molekulare Diagnostik



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PPP2R5E siRNA (m): sc-152427



The Power to Question

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. PPP2R5E (protein phosphatase 2, regulatory subunit B', ϵ isoform) is a 467 amino acid protein that localizes to the cytoplasm and exists as an isoform of the B regulatory subunit within the PP multimeric complex. Functioning as a regulatory subunit, PPP2R5E is thought to modulate both the catalytic activity and the substrate specificity of the PP holoenzyme and may also be responsible for the localization of the complex to subcellular compartments.

REFERENCES

- 1. McCright, B., et al. 1995. Identification of a new family of protein phosphatase 2A regulatory subunits. J. Biol. Chem. 270: 26123-26128.
- 2. McCright, B., et al. 1996. Assignment of human protein phosphatase 2A regulatory subunit genes $b56\alpha$, $b56\beta$, $b56\gamma$, $b56\delta$, and $b56\varepsilon$ (PPP2R5A-PPP2R5E), highly expressed in muscle and brain, to chromosome regions 1q41, 11q12, 3p21, 6p21.1, and 7p11.2 \rightarrow p12. Genomics 36: 168-170.
- 3. McCright, B., et al. 1996. The B56 family of protein phosphatase 2A (PP2A) regulatory subunits encodes differentiation-induced phosphoproteins that target PP2A to both nucleus and cytoplasm. J. Biol. Chem. 271: 22081-22089.
- Dozier, C., et al. 2004. Regulation of Chk2 phosphorylation by interaction with protein phosphatase 2A via its B' regulatory subunit. Biol. Cell 96: 509-517.
- Ammosova, T., et al. 2005. Dephosphorylation of CDK9 by protein phosphatase 2A and protein phosphatase-1 in Tat-activated HIV-1 transcription. Retrovirology 2: 47.
- 6. Sablina, A.A., et al. 2007. The tumor suppressor PP2A A β regulates the RaIA GTPase. Cell 129: 969-982.

CHROMOSOMAL LOCATION

Genetic locus: Ppp2r5e (mouse) mapping to 12 C3.

PRODUCT

PPP2R5E 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 PPP2R5E shRNA Plasmid (m): sc-152427-SH and PPP2R5E shRNA (m) Lentiviral Particles: sc-152427-V as alternate gene silencing products.

For independent verification of PPP2R5E (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-152427A, sc-152427B and sc-152427C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

PPP2R5E siRNA (m) is recommended for the inhibition of PPP2R5E 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

PPP2R5E (A-11): sc-376176 is recommended as a control antibody for monitoring of PPP2R5E 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-lgG κ BP-HRP: sc-516102 or m-lgG 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-lgG κ BP-FITC: sc-516140 or m-lgG 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 PPP2R5E gene expression knockdown using RT-PCR Primer: PPP2R5E (m)-PR: sc-152427-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.

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