

I2PP2A siRNA (r): sc-106998

BACKGROUND

Protein phosphatase 2A PP2A is a major mammalian protein serine/threonine phosphatase that regulates diverse cellular processes. Inhibitor 1 of PP2A (I1PP2A) and inhibitor 2 of PP2A (I2PP2A), which share large sequence similarity, are heat-stable protein inhibitors of the cellular phosphatase activity of PP2A. I1PP2A and I2PP2A were initially characterized as putative HLA class II associated proteins Phap I and Phap II. These inhibitor proteins act noncompetitively to selectively inhibit PP2A, but do not affect the phosphatase activity of the related proteins PP1, PP2B and PP2C. The I1PP2A protein is localized to both the cytoplasm and the nucleus. In contrast, I2PP2A is located predominantly in the nucleus and is highly expressed in Wilms' tumor cells. Transient expression of I2PP2A in HEK-293 cells leads to an increase in the DNA binding activity of the proto-oncogene c-Jun.

REFERENCES

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- Li, M., et al. 1995. Purification and characterization of two potent heat-stable protein inhibitors of protein phosphatase 2A from bovine kidney. *Biochemistry* 34: 1988-1996.
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- Carlson, S.G., et al. 1998. Expression of SET, an inhibitor of protein phosphatase 2A, in renal development and Wilms' tumor. *J. Am. Soc. Nephrol.* 9: 1873-1880.

CHROMOSOMAL LOCATION

Genetic locus: Set (rat) mapping to 19q12.

PRODUCT

I2PP2A siRNA (r) 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 I2PP2A shRNA Plasmid (r): sc-106998-SH and I2PP2A shRNA (r) Lentiviral Particles: sc-106998-V as alternate gene silencing products.

For independent verification of I2PP2A (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-106998A, sc-106998B and sc-106998C.

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

I2PP2A siRNA (r) is recommended for the inhibition of I2PP2A 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

I2PP2A (F-9): sc-133138 is recommended as a control antibody for monitoring of I2PP2A 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 I2PP2A gene expression knockdown using RT-PCR Primer: I2PP2A (r)-PR: sc-106998-PR (20 μ l, 582 bp). 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.