Wip1 siRNA (m): sc-39206



The Power to Question

BACKGROUND

Several major protein serine/threonine phosphatases have been identified in eukaryotic cells. These include protein phosphatase families 1, 2A, 2B, 2C, X and Y (PP-1, PP-2A, PP-2B, PP-2C, PP-X and PP-Y). These enzymes can be distinguished by their action on phosphorylase kinase and their sensitivity to certain activators and inhibitors. Wip1 (wildtype p53-induced phosphatase or PPM1D), a protein identified in the p53 DNA response pathway, is a member of the PP2C family. Wip1 is a serine/threonine protein phosphatase which selectively inactivates p38 MAPK and dephosphorylates the ATM/ATR targets, Chk1 and p53. Wip1 is ubiquitously expressed but is present at very high levels in testis. Deletion of Wip1 results in a reduction of T and B cell function and compromised cell division, rendering cells resistant to becoming cancerous and slowing tumor development.

REFERENCES

- 1. Ingebritsen, T.S., et al. 1983. Protein phosphatases: properties and role in cellular regulation. Science 221: 331-338.
- Cohen, P. 1989. The structure and regulation of protein phosphatases. Annu. Rev. Biochem. 58: 453-508.
- 3. Cohen, P.T., et al. 1990. Protein serine/threonine phosphatases: an expanding family. FEBS Lett. 268: 335-359.
- Kamibayashi, C., et al. 1991. Subunit interactions control protein phosphatase 2A. Effects of limited proteolysis, N-ethylmaleimide, and heparin on the interaction of the β subunit. J. Biol. Chem. 266: 13251-13260.
- Shenolikar, S., et al. 1991. Protein phosphatases: recent progress. Adv. Second Messenger Phosphoprotein Res. 23: 1-121.
- Fiscella, M., et al. 1997. Wip1, a novel human protein phosphatase that is induced in response to ionizing radiation in a p53-dependent manner. Proc. Natl. Acad. Sci. USA 94: 6048-6053.
- Shreeram, S., et al. 2006. Regulation of Atm/p53-dependent suppression of Myc-induced lymphomas by Wip1 phosphatase. J. Exp. Med. 203: 2793-2799.
- 8. Schito, M.L., et al. 2006. Wip1 phosphatase-deficient mice exhibit defective T cell maturation due to sustained p53 activation. J. Immunol. 176: 4818-4825.

CHROMOSOMAL LOCATION

Genetic locus: Ppm1d (mouse) mapping to 11 C.

PRODUCT

Wip1 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 Wip1 shRNA Plasmid (m): sc-39206-SH and Wip1 shRNA (m) Lentiviral Particles: sc-39206-V as alternate gene silencing products.

For independent verification of Wip1 (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-39206A, sc-39206B and sc-39206C.

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

Wip1 siRNA (m) is recommended for the inhibition of Wip1 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

Wip1 (F-10): sc-376257 is recommended as a control antibody for monitoring of Wip1 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 Wip1 gene expression knockdown using RT-PCR Primer: Wip1 (m)-PR: sc-39206-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.