SANTA CRUZ BIOTECHNOLOGY, INC.

PME-1 siRNA (h): sc-36281

BACKGROUND

Protein phosphatase methylesterase-1 (PME-1) catalyzes the demethylation and inactivation of protein phosphatase (PP2A), which is a multimeric phosphoserine/threonine protein phosphatase associated with growth inhibition and cell cycle arrest. Carboxymethylation and demethylation is a covalent modifcation that regulates the catalytic activity of certain proteins in eukaryotes. Electrostatic interactions that occur at residues or metals in or near the active site can influence the specificity of carboxymethylation and demethylation. PME-1 can demethylate PP2A catalytic subunit *in vitro* and okadaic acid treatment is capable of inhibiting this reaction. PME-1 is conserved from yeast to human and contains a motif found in lipases having a catalytic triad-activated serine as their active site nucleophile.

REFERENCES

1. Lee, J., et al. 1996. A specific protein carboxyl methylesterase that demethylates phosphoprotein phosphatase 2A in bovine brain. Proc. Natl. Acad. Sci. USA 93: 6043-6047.
2. Schonthal, A.H. 1998. Role of PP2A in intracellular signal transduction pathways. Front. Biosci. 3: D1262-D1273.
3. Ogris, E., et al. 1999. A protein phosphatase methylesterase (PME-1) is one of several novel proteins stably associating with two inactive mutants of protein phosphatase 2A. J. Biol. Chem. 274: 14382-14391.
4. Wu, J., et al. 2000. Carboxyl methylation of the phosphoprotein phosphatase 2A catalytic subunit promotes its functional association with regulatory subunits *in vivo*. EMBO J. 19: 5672-5681.
5. Tolstykh, T., et al. 2000. Carboxyl methylation regulates phosphoprotein phosphatase 2A by controlling the association of regulatory B subunits. EMBO J. 19: 5682-5691.
6. Gagnon, S.N., et al. 2002. The genes PME-1 and PME-2 encode two poly (ADP-ribose) polymerases in *C. elegans*. Biochem. J. 368: 263-271.
7. Longin, S., et al. 2004. An inactive protein phosphatase 2A population is associated with methyl-esterase and can be re-activated by the phosphotyrosyl phosphatase activator. Biochem. J. 380: 111-119.

CHROMOSOMAL LOCATION

Genetic locus: PPME1 (human) mapping to 11q13.4.

PRODUCT

PME-1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PME-1 shRNA Plasmid (h): sc-36281-SH and PME-1 shRNA (h) Lentiviral Particles: sc-36281-V as alternate gene silencing products.

For independent verification of PME-1 (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-36281A, sc-36281B and sc-36281C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PME-1 siRNA (h) is recommended for the inhibition of PME-1 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 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

PME-1 (B-12): sc-25278 is recommended as a control antibody for monitoring of PME-1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 PME-1 gene expression knockdown using RT-PCR Primer: PME-1 (h)-PR: sc-36281-PR (20 µl). 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.