

PMF-1 siRNA (h): sc-78780

BACKGROUND

PMF-1 (polyamine-modulated factor 1) is a 205 amino acid protein involved in kinetochore formation. Localized to the nucleus, PMF-1 contains a coiled-coil domain which interacts with the leucine-zipper domain of Nrf2. This interaction regulates the transcription of SSAT, a regulatory enzyme for polyamine catabolism. PMF-1 is also, a component of the MIS12 complex, which is required for kinetochore formation and chromosomal alignment and segregation. PMF-1 is expressed at highest levels in skeletal muscle and heart, with moderate expression in liver and kidney. PMF-1 exists as five isoforms produced by alternative splicing.

REFERENCES

1. Wang, Y., et al. 1999. Cloning and characterization of human polyamine-modulated factor-1, a transcriptional cofactor that regulates the transcription of the spermidine/spermine N¹-acetyltransferase gene. *J. Biol. Chem.* 274: 22095-22101.
2. Wang, Y., et al. 2001. Characterization of the interaction between the transcription factors human polyamine modulated factor (PMF-1) and NF-E2-related factor 2 (Nrf-2) in the transcriptional regulation of the spermidine/spermine N¹-acetyltransferase (SSAT) gene. *Biochem. J.* 355: 45-49.
3. Wang, Y., et al. 2002. Polyamine-modulated factor 1 binds to the human homologue of the 7a subunit of the *Arabidopsis* COP9 signalosome: implications in gene expression. *Biochem. J.* 366: 79-86.
4. Cheeseman, I.M., et al. 2004. A conserved protein network controls assembly of the outer kinetochore and its ability to sustain tension. *Genes Dev.* 18: 2255-2268.
5. Obuse, C., et al. 2004. A conserved Mis12 centromere complex is linked to heterochromatic HP1 and outer kinetochore protein Zwint-1. *Nat. Cell Biol.* 6: 1135-1141.
6. Kline, S.L., et al. 2006. The human Mis12 complex is required for kinetochore assembly and proper chromosome segregation. *J. Cell Biol.* 173: 9-17.
7. Hyvönen, M.T., et al. 2006. Polyamine-regulated unproductive splicing and translation of spermidine/spermine N¹-acetyltransferase. *RNA* 12: 1569-1582.

CHROMOSOMAL LOCATION

Genetic locus: PMF1 (human) mapping to 1q22.

PRODUCT

PMF-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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PMF-1 shRNA Plasmid (h): sc-78780-SH and PMF-1 shRNA (h) Lentiviral Particles: sc-78780-V as alternate gene silencing products.

For independent verification of PMF-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-78780A, sc-78780B and sc-78780C.

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

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

PMF-1 (B-5): sc-376988 is recommended as a control antibody for monitoring of PMF-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-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 PMF-1 gene expression knockdown using RT-PCR Primer: PMF-1 (h)-PR: sc-78780-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.