

PRDM16 siRNA (h): sc-62854

BACKGROUND

The PR-domain containing proteins (PRDMs) have a common involvement in the modulation of gene activities. A PR-domain family member usually produces two products, called PR-plus and PR-minus, which differ by the presence or absence of the PR domain, respectively. The PR-plus product is underexpressed or disrupted in cancer cells, whereas the PR-minus product is present or overexpressed in cancer cells. This imbalance in the amount of the two products, which is a result of either genetic or epigenetic events, appears to be a determining factor of malignancy. PRDM16 (PR domain containing 16), also known as MEL1 or PFM13, is a 1,276 amino acid protein that contains one SET domain and ten C₂H₂-type zinc fingers. Localized to the nucleus, PRDM16 functions as a transcription factor and is thought to be involved in the pathogenesis of acute myeloid leukemia and myelodysplastic syndrome. Three isoforms of PRDM16 exist due to alternative splicing events.

REFERENCES

1. Mochizuki, N., et al. 2000. A novel gene, MEL1, mapped to 1p36.3 is highly homologous to the MDS1/EVI1 gene and is transcriptionally activated in t(1;3)(p36;q21)-positive leukemia cells. *Blood* 96: 3209-3214.
2. Nishikata, I., et al. 2003. A novel EVI1 gene family, MEL1, lacking a PR domain (MEL1S) is expressed mainly in t(1;3)(p36;q21)-positive AML and blocks G-CSF-induced myeloid differentiation. *Blood* 102: 3323-3332.
3. Xinh, P.T., et al. 2003. Breakpoints at 1p36.3 in three MDS/AML(M4) patients with t(1;3)(p36;q21) occur in the first intron and in the 5' region of MEL1. *Genes Chromosomes Cancer* 36: 313-316.
4. Lahortiga, I., et al. 2004. Molecular characterization of a t(1;3)(p36;q21) in a patient with MDS. MEL1 is widely expressed in normal tissues, including bone marrow, and it is not overexpressed in the t(1;3) cells. *Oncogene* 23: 311-316.
5. Ott, M.G., et al. 2006. Correction of X-linked chronic granulomatous disease by gene therapy, augmented by insertional activation of MDS1-EVI1, PRDM16 or SETBP1. *Nat. Med.* 12: 401-409.

CHROMOSOMAL LOCATION

Genetic locus: PRDM16 (human) mapping to 1p36.32.

PRODUCT

PRDM16 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 PRDM16 shRNA Plasmid (h): sc-62854-SH and PRDM16 shRNA (h) Lentiviral Particles: sc-62854-V as alternate gene silencing products.

For independent verification of PRDM16 (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-62854A, sc-62854B and sc-62854C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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

PRDM16 siRNA (h) is recommended for the inhibition of PRDM16 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

PRDM16 (174A2D): sc-517625 is recommended as a control antibody for monitoring of PRDM16 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 PRDM16 gene expression knockdown using RT-PCR Primer: PRDM16 (h)-PR: sc-62854-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.