

MLF1 siRNA (m): sc-61056

BACKGROUND

Myeloid leukemia factor 1 (MLF1) is a 268 amino acid protein expressed by a gene that is involved in translocations associated with acute myeloid leukemia. MLF1 is a widely expressed negative regulator of cell cycle progression functioning upstream of the tumor suppressor p53. MLF1 induces p53-dependent cell cycle arrest in murine embryonic fibroblasts. MLF1 expression also inversely affects the endogenous level of COP1, a ubiquitin ligase for p53, inhibits Epo-induced cell cycle exit and inhibits a rise in the cell cycle inhibitor p27. Polo-like kinase 1 (Plk1) phosphorylates MLF1 at its Thr 78 site, which induces ubiquitination and degradation of MLF1 before the transition from metaphase to anaphase. Mutations of these phosphorylation sites stabilize MLF1 and inhibit mitotic progression. MLF1 normally functions in multipotent progenitor cells and its dysregulation may be somewhat responsible for leukemogenesis.

REFERENCES

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3. Kazemi-Esfarjani, P. and Benzer, S. 2002. Suppression of polyglutamine toxicity by a *Drosophila* homolog of myeloid leukemia factor 1. *Hum. Mol. Genet.* 11: 2657-2672.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 601402. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Hanissian, S.H., et al. 2004. cDNA cloning and characterization of a novel gene encoding protein MLF1IP. *Oncogene* 23: 3700-3707.
6. Winteringham, L.N., et al. 2004. Myeloid Leukemia Factor 1 inhibits erythropoietin-induced differentiation, cell cycle exit and p27 Kip1 accumulation. *Oncogene* 23: 5105-5109.
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CHROMOSOMAL LOCATION

Genetic locus: Mlf1 (mouse) mapping to 3 E1.

PRODUCT

MLF1 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 MLF1 shRNA Plasmid (m): sc-61056-SH and MLF1 shRNA (m) Lentiviral Particles: sc-61056-V as alternate gene silencing products.

For independent verification of MLF1 (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-61056A, sc-61056B and sc-61056C.

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

MLF1 siRNA (m) is recommended for the inhibition of MLF1 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 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

MLF1 (D-1): sc-514294 is recommended as a control antibody for monitoring of MLF1 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 MLF1 gene expression knockdown using RT-PCR Primer: MLF1 (m)-PR: sc-61056-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.