

Mcl-1 siRNA (m): sc-35878

BACKGROUND

B cell CLL/lymphoma-2 (Bcl-2) blocks cell death following a variety of stimuli and confers a death-sparing effect to certain hematopoietic cell lines following growth factor withdrawal. Myeloid cell leukemia-1 (Mcl-1) shares sequence homology with Bcl-2 and further resembles Bcl-2 in that its expression promotes cell viability. p53 and Mcl-1 demonstrate opposing effects on mitochondrial apoptosis by mediating Bcl-2 antagonist killer (Bak) activity. Mcl-1 is an important and specific regulator that is necessary for the homeostasis of early hematopoietic progenitors. Glycogen synthase kinase-3 (GSK-3) controls Mcl-1 stability, which has an effect on the regulation of apoptosis by growth factors PI 3-kinase and Akt. Mice with a deficiency of the Mcl-1 protein show a significant reduction in B and T lymphocytes similar to the effects observed in IL-7- or IL-7R-deficient mice.

REFERENCES

1. Kozopas, K.M., et al. 1993. Mcl-1, a gene expressed in programmed myeloid cell differentiation, has sequence similarity to Bcl-2. *Proc. Natl. Acad. Sci. USA* 90: 3516-3520.
2. Craig, R.W., et al. 1994. Human and mouse chromosomal mapping of the myeloid cell leukemia-1 gene: Mcl-1 maps to human chromosome 1q21, a region that is frequently altered in preneoplastic and neoplastic disease. *Genomics* 23: 457-463.
3. Rinkenberger, J.L., et al. 2000. Mcl-1 deficiency results in peri-implantation embryonic lethality. *Genes Dev.* 14: 23-27.
4. Bae, J., et al. 2000. Mcl-1S, a splicing variant of the anti-apoptotic Bcl-2 family member Mcl-1, encodes a pro-apoptotic protein possessing only the BH3 domain. *J. Biol. Chem.* 275: 25255-25261.

CHROMOSOMAL LOCATION

Genetic locus: Mcl1 (mouse) mapping to 3 F2.1.

PRODUCT

Mcl-1 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 Mcl-1 shRNA Plasmid (m): sc-35878-SH and Mcl-1 shRNA (m) Lentiviral Particles: sc-35878-V as alternate gene silencing products.

For independent verification of Mcl-1 (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-35878A, sc-35878B and sc-35878C.

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

Mcl-1 siRNA (m) is recommended for the inhibition of Mcl-1 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

Mcl-1 (B-6): sc-74436 is recommended as a control antibody for monitoring of Mcl-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 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 Mcl-1 gene expression knockdown using RT-PCR Primer: Mcl-1 (m)-PR: sc-35878-PR (20 μ l, 433 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Breitenbuecher, F., et al. 2009. A novel molecular mechanism of primary resistance to FLT3-kinase inhibitors in AML. *Blood* 113: 4063-4073.
2. Eno, C.O., et al. 2012. The Bcl-2 proteins Noxa and Bcl-x_L co-ordinately regulate oxidative stress-induced apoptosis. *Biochem. J.* 444: 69-78.
3. Giri, J., et al. 2016. *Leishmania donovani* exploits myeloid cell leukemia 1 (Mcl-1) protein to prevent mitochondria-dependent host cell apoptosis. *J. Biol. Chem.* 291: 3496-3507.
4. Giri, J., et al. 2022. Translationally controlled tumor protein-mediated stabilization of host antiapoptotic protein Mcl-1 is critical for establishment of infection by intramacrophage parasite *Leishmania donovani*. *J. Immunol.* 208: 2540-2548.

RESEARCH USE

For research use only, not for use in diagnostic procedures.