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B-Myb siRNA (h): sc-43523

BACKGROUND

The highly leukemogenic avian retrovirus E26 contains two oncogenes, v-Myb and v-Ets, which are expressed together as a fusion protein. The cellular homolog of v-Myb, designated c-Myb, encodes a transcription factor. Deletion or disruption of a negative regulatory domain mapping within the carboxy terminal domain of c-Myb results in enhanced transactivating capacity and in parallel, leads to activation of its ability to transform hemopoietic cells. c-Myb is expressed preferentially, but not exclusively, in immature hemopoietic cells and its expression decreases as cells differentiate. A second member of the Myb proto-oncogene family, B-Myb, encodes a second sequence-specific DNA binding protein. B-Myb RNA levels are low or undetectable in quiescent cells but increase at the G₁/S-phase transition following mitogenic stimulation. Studies suggest that B-Myb expression rescues cells from p53-induced G₁ arrest mediated by p21.

REFERENCES

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- Sakura, H., et al. 1989. Delineation of three functional domains of the transcriptional activator encoded by the c-Myb protooncogene. *Proc. Natl. Acad. Sci. USA* 86: 5758-5762.
- Mizuguchi, G., et al. 1990. DNA binding activity and transcriptional activator function of the human B-Myb protein compared with c-Myb. *J. Biol. Chem.* 265: 9280-9284.
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- Lin, D., et al. 1994. Constitutive expression of B-Myb can bypass p53-induced Waf1/Cip1-mediated G₁ arrest. *Proc. Natl. Acad. Sci. USA* 91: 10079-10083.

CHROMOSOMAL LOCATION

Genetic locus: MYBL2 (human) mapping to 20q13.12.

PRODUCT

B-Myb 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 B-Myb shRNA Plasmid (h): sc-43523-SH and B-Myb shRNA (h) Lentiviral Particles: sc-43523-V as alternate gene silencing products.

For independent verification of B-Myb (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-43523A, sc-43523B and sc-43523C.

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

B-Myb siRNA (h) is recommended for the inhibition of B-Myb 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

B-Myb (C-5): sc-390198 is recommended as a control antibody for monitoring of B-Myb 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 B-Myb gene expression knockdown using RT-PCR Primer: B-Myb (h)-PR: sc-43523-PR (20 μ l, 575 bp). 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.