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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

linkedin.com/company/szaboscandic

SANTA CRUZ BIOTECHNOLOGY, INC.

B-Myb shRNA (m) Lentiviral Particles: sc-43524-V

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 G_1/S-phase transition following mitogenic stimulation. Studies suggest that B-Myb expression rescues cells from p53-induced G_1 arrest mediated by p21.

REFERENCES

1. Gonda, T.J., et al. 1984. Expression of myb, myc and fos proto-oncogenes during the differentiation of a murine myeloid leukaemia. Nature 310: 249-251.
2. Gonda, T.J., et al. 1985. Nucleotide sequence of cDNA clones of the murine myb proto-oncogene. EMBO J. 4: 2004-2008.
3. 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.
4. 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.
5. Ramsay, R.G., et al. 1991. Increase in specific DNA binding by carboxyl truncation suggests a mechanism for activation of Myb. Oncogene 6: 1875-1879.
6. Favier, D., et al. 1994. Detection of proteins that bind to the leucine zipper motif of c-Myb. Oncogene. 9: 305-311.

CHROMOSOMAL LOCATION

Genetic locus: Mybl2 (mouse) mapping to 2 H2.

PRODUCT

B-Myb shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see B-Myb siRNA (m): sc-43524 and B-Myb shRNA Plasmid (m): sc-43524-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

B-Myb shRNA (m) Lentiviral Particles is recommended for the inhibition of B-Myb expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

B-Myb (C-20): sc-725 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor B-Myb gene expression knockdown using RT-PCR Primer: B-Myb (m)-PR: sc-43524-PR (20 µl, 476 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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