SANTA CRUZ BIOTECHNOLOGY, INC.

MIB1 siRNA (h): sc-75781

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The Power to Question

BACKGROUND

The LIN-12/notch family of transmembrane receptors is believed to play a central role in development by regulating cell fate decisions. MIB1 (E3 ubiquitin-protein ligase MIB1), also known as mind bomb homolog 1 and DAPK-interacting protein 1, is a 1,006 amino acid E3 ubiquitin ligase that activates the notch ligand Delta. MIB1 ubiquinates Delta by binding to its intracellular domain, leading to the endocytosis and eventual degradation of the Delta receptor, which, paradoxically, results in the up-regulation of receptor activity and enhances notch signaling. MIB1 also interacts with DAPK, a protein that plays an important role in the regulation of apoptosis. Ubiquination of DAPK leads to inhibition of caspase-dependent apoptosis, therefore it is likely that overexpression of MIB1 can lead to tumor growth. Although it seems to be widely expressed at low levels, MIB1 is expressed at highest concentrations in the CNS and ovary. Both DAPK and MIB1 are overexpressed in epileptic brain tissue, suggesting that they probably cooperate as regulators of neuronal death in epilepsy.

REFERENCES

1. Jin, Y., et al. 2002. A death-associated protein kinase (DAPK)-interacting protein, DIP-1, is an E3 ubiquitin ligase that promotes tumor necrosis factor-induced apoptosis and regulates the cellular levels of DAPK. J. Biol. Chem. 277: 46980-46986.
2. Le Borgne, R. and Schweisguth, F. 2003. Notch signaling: endocytosis makes Delta signal better. Curr. Biol. 13: R273-R275.
3. Itoh, M., et al. 2003. Mind bomb is a ubiquitin ligase that is essential for efficient activation of Notch signaling by Delta. Dev. Cell 4: 67-82.
4. Henshall, D.C., et al. 2004. Death-associated protein kinase expression in human temporal lobe epilepsy. Ann. Neurol. 55: 485-494.
5. Online Mendelian Inheritance in Man, OMIM™. 2005. Johns Hopkins University, Baltimore, MD. MIM Number: 608677. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Choe, E.A., et al. 2007. Neuronal morphogenesis is regulated by the interplay between cyclin-dependent kinase 5 and the ubiquitin ligase mind bomb 1. J. Neurosci. 27: 9503-9512.

CHROMOSOMAL LOCATION

Genetic locus: MIB1 (human) mapping to 18q11.2.

PRODUCT

MIB1 siRNA (h) is a target-specific 19-25 nt siRNA designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MIB1 shRNA Plasmid (h): sc-75781-SH and MIB1 shRNA (h) Lentiviral Particles: sc-75781-V as alternate gene silencing products.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

MIB1 siRNA (h) is recommended for the inhibition of MIB1 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 µM in 66 µl. 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

MIB1 (D-6): sc-393551 is recommended as a control antibody for monitoring of MIB1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 MIB1 gene expression knockdown using RT-PCR Primer: MIB1 (h)-PR: sc-75781-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Ray, J., et al. 2019. MicroRNA-198 suppresses prostate tumorigenesis by targeting MIB1. Oncol. Rep. 42: 1047-1056.

RESEARCH USE

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