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SANTA CRUZ BIOTECHNOLOGY, INC.

Nmi siRNA (h2): sc-156150

BACKGROUND

Nmi (for N-Myc interactor) is an interferon inducible protein that associates with multiple transcription factors, including c-Myc, n-Myc, Max, and c-Fos, which contain bHLH-ZIP, bHLH, or Zip domains. Nmi is ubiquitously expressed at low levels throughout various fetal and adult tissues and at higher levels in myeloid leukemias and cell lines overexpressing c-Myc. In addition to binding Myc proteins, Nmi also associates with the Stat family of transcription factors, where it enhances Stat-dependent transcription. Although Nmi lacks an intrinsic DNA binding or activation domain, Nmi enhances the transcriptional activity of the Stat proteins, in response to cytokine stimulation, by recruiting the Stat1 and Stat5 transcriptional coactivators, CREB-binding protein (CBP) and p300. *In vitro* studies indicate that Nmi, expressed in conjunction with CBP, enhances the transcriptional responsiveness of Stat5 to IL-2 stimulation five fold over CBP alone by increasing the affinity of Stat proteins for CBP/p300.

REFERENCES

1. Bao, J., et al. 1996. Isolation and characterization of Nmi, a novel partner of Myc proteins. Oncogene 12: 2171-2176.
2. Lebrun, S.J., et al. 1998. Interferon-induced upregulation and cytoplasmic localization of Myc-interacting protein Nmi. J. Interferon Cytokine Res. 18: 767-771.
3. Lee, N.D., et al. 1999. Subcellular localization of interferon-inducible Myc/Stat-interacting protein Nmi is regulated by a novel IFP 35 homologous domain. J. Interferon Cytokine Res. 19: 1245-1252.
4. Gingras, S., et al. 1999. p300/CBP is required for transcriptional induction by interleukin-4 and interacts with Stat6. Nucleic Acids Res. 27: 2722-2729.
5. Sakamuro, D., et al. 1999. New Myc-interacting proteins: a second Myc network emerges. Oncogene 18: 2942-2954.
6. Zhu, M., et al. 1999. Functional association of Nmi with Stat5 and Stat1 in IL-2- and IFNγ-mediated signaling. Cell 96: 121-130.
7. Li, H., et al. 2002. A novel tricomplex of BRCA1, Nmi, and c-Myc inhibits c-Myc-induced human telomerase reverse transcriptase gene (hTERT) promoter activity in breast cancer. J. Biol. Chem. 277: 20965-20973.

CHROMOSOMAL LOCATION

Genetic locus: NMI (human) mapping to 2q23.3.

PRODUCT

Nmi siRNA (h2) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Nmi shRNA Plasmid (h2): sc-156150-SH and Nmi shRNA (h2) Lentiviral Particles: sc-156150-V as alternate gene silencing products.

For independent verification of Nmi (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-156150A, sc-156150B and sc-156150C.

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

Nmi siRNA (h2) is recommended for the inhibition of Nmi 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

Nmi (D-10): sc-377177 is recommended as a control antibody for monitoring of Nmi 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 Nmi gene expression knockdown using RT-PCR Primer: Nmi (h2)-PR: sc-156150-PR (20 µl). 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.