SANTA CRUZ BIOTECHNOLOGY, INC.

N4BP1 siRNA (h): sc-92981

BACKGROUND

N4BP1 (NEDD4 binding protein 1) is an 896 amino acid protein that belongs to the N4BP1 family. N4BP1 primarily localizes to nucleolus, however it also localizes to PML nuclear bodies when desumoylated. Detected in heart, lung, brain, liver, skeletal muscle, pancreas, kidney, spleen, testis and ovary, N4BP1 is phosphorylated upon DNA damage probably by ATM or ATR. N4BP1 interacts with Nedd4, a protein with a wide range of activities including modification of ubiquitin-protein ligase, regulation of membrane channels and potentiating hormone-dependent activation of transcription. N4BP1 inhibits the E3 ubiquitin-protein ligase ITCH, by interacting with the second WW domain of ITCH thereby competing with ITCH's substrates and impairing ubiquitination of substrates. N4BP1 has no obvious transmembrane regions, which raises the possibility that N4BP1 represents a soluble adaptor that could sequester ITCH within the cytosol. The N4BP1 gene maps to chromosome 16q12.1.

REFERENCES

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3. Melino, G., et al. 2008. Itch: a HECT-type E3 ligase regulating immunity, skin and cancer. Cell Death Differ. 15: 1103-1112.
4. Gay, D.L., et al. 2008. Cbl- and Nedd4-family ubiquitin ligases: balancing tolerance and immunity. Immunol. Res. 42: 51-64.
5. Levy, D., et al. 2008. A regulatory circuit controlling Itch-mediated p73 degradation by Runx. J. Biol. Chem. 283: 27462-27468.
6. Rotin, D. and Kumar, S. 2009. Physiological functions of the HECT family of ubiquitin ligases. Nat. Rev. Mol. Cell. Biol. 10: 398-409.
7. Sharma, P., et al. 2010. N4BP1 is a newly identified nucleolar protein that undergoes SUMO-regulated polyubiquitylation and proteasomal turnover at promyelocytic leukemia nuclear bodies. J. Cell Sci. 123: 1227-1234.

CHROMOSOMAL LOCATION

Genetic locus: N4BP1 (human) mapping to 16q12.1.

PRODUCT

N4BP1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see N4BP1 shRNA Plasmid (h): sc-92981-SH and N4BP1 shRNA (h) Lentiviral Particles: sc-92981-V as alternate gene silencing products.

For independent verification of N4BP1 (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-92981A, sc-92981B and sc-92981C.

RESEARCH USE

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

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

N4BP1 siRNA (h) is recommended for the inhibition of N4BP1 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor N4BP1 gene expression knockdown using RT-PCR Primer: N4BP1 (h)-PR: sc-92981-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.