SANTA CRUZ BIOTECHNOLOGY, INC.

nucleobindin siRNA (m): sc-40779

BACKGROUND

Nucleobindin, also designated Nuc or Calnuc, is a secreted protein that promotes production of DNA-specific antibodies in lupus prone MRL/lpr mice. Nucleobindin contains a signal peptide, two EF-hand motifs, acidic and basic regions and a leucine-zipper motif. Nucleobindin has two calcium-binding domains and is the major Golgi Ca^{2+} binding protein. The leucine zipper structure and the basic amino acid-rich region are responsible for DNA binding. Nucleobindin preferentially associates with membranes of polarized cells. Nucleobindin is found in both the cytosol and the membrane and is localized to *cis*-Golgi cisternae and the *cis*-Golgi network (CGN). Nucleobindin is involved in autoimmunity, apoptosis and calcium homeostasis in the bone matrix.

REFERENCES

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2. Miura, K., et al. 1996. Organization of the human gene for nucleobindin (NUC) and its chromosomal assignment to 19q13.2-q13.4. Genomics 34: 181-186.
3. Kubota, T., et al. 1998. Upregulation of nucleobindin expression in human-activated lymphocytes and non-Hodgkin's lymphoma. Pathol. Int. 48: 22-28.
4. Lin, P., et al. 1998. The mammalian calcium-binding protein, nucleobindin (CALNUC), is a Golgi resident protein. J. Cell Biol. 141: 1515-1527.
5. Fialka, I., et al. 1999. Loss of epithelial polarity is accompanied by differential association of proteins with intracellular membranes. Electrophoresis 20: 331-343.
6. Lin, P., et al. 1999. Overexpression of CALNUC (nucleobindin) increases agonist and thapsigargin releasable Ca^2 storage in the Golgi. J. Cell Biol. 145: 279-289.
7. Kawano, J., et al. 2000. CALNUC (nucleobindin) is localized in the Golgi apparatus in insect cells. Eur. J. Cell Biol. 79: 208-217.
8. Lin, P., et al. 2000. Calnuc, an EF-hand Ca^{2+} binding protein, specifically interacts with the C-terminal α5-helix of $G_{\alpha\ 1\text{-}3}$. Proc. Natl. Acad. Sci. USA 97: 674-679.

CHROMOSOMAL LOCATION

Genetic locus: Nucb1 (mouse) mapping to 7 B3.

PRODUCT

nucleobindin siRNA (m) 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 nucleobindin shRNA Plasmid (m): sc-40779-SH and nucleobindin shRNA (m) Lentiviral Particles: sc-40779-V as alternate gene silencing products.

For independent verification of nucleobindin (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-40779A, sc-40779B and sc-40779C.

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

nucleobindin siRNA (m) is recommended for the inhibition of nucleobindin expression in mouse 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

nucleobindin (D-8): sc-515246 is recommended as a control antibody for monitoring of nucleobindin 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 nucleobindin gene expression knockdown using RT-PCR Primer: nucleobindin (m)-PR: sc-40779-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.