SANTA CRUZ BIOTECHNOLOGY, INC.

nucleobindin 2 siRNA (h): sc-62705

BACKGROUND

Nucleobindin 2, also designated NUCB2 or NEFA, is a 420 amino acid protein that is predominantly expressed in spleen, testis and stomach. It localizes to the Golgi and cisternae of the endoplasmic reticulum (ER) and in the nuclear envelope of neurons in the brain. Nucleobindin 2 contains leucine-zipper and EF-hand motifs, two helix-loop-helix regions, and both a basic and an acidic amino acid region. The leucine zipper structure and the basic amino acid-rich region are responsible for DNA binding. It is a highly charged protein that binds Ca^{2+} via its EF-hand domains. Nucleobindin 2 is also expressed in the hypothalamic nuclei in rats, which may indicate a role in appetite control. Conversion of nucleobindin 2 to nesfatin-1 in the brain decreases food intake in rats. Nesfatin-1 is identified as a satiety molecule that is involved in melanocortin signaling in the hypothalamus.

REFERENCES

1. Barnikol-Watanabe, S., et al. 1994. Human protein NEFA, a novel DNA binding/EF-hand/leucine zipper protein. Molecular cloning and sequence analysis of the cDNA, isolation and characterization of the protein. Biol. Chem. Hoppe Seyler 375: 497-512.
2. Kroll, K.A., et al. 1999. Heterologous overexpression of human NEFA and studies on the two EF-hand calcium-binding sites. Biochem. Biophys. Res. Commun. 260: 1-8.
3. Taniguchi, N., et al. 2000. The postmitotic growth suppressor necdin interacts with a calcium-binding protein (NEFA) in neuronal cytoplasm. J. Biol. Chem. 275: 31674-31681.
4. Caldwell, G.M., et al. 2001. Mapping of genes and transcribed sequences in a gene rich 400-kb region on human chromosome 11p15.1→p14. Cytogenet. Cell Genet. 92: 103-107.
5. Nesselhut, J., et al. 2001. Golgi retention of human protein NEFA is mediated by its N-terminal Leu/Ile-rich region. FEBS Lett. 509: 469-475.
6. Oh-I, S., et al. 2006. Identification of nesfatin-1 as a satiety molecule in the hypothalamus. Nature 443: 709-712.

CHROMOSOMAL LOCATION

Genetic locus: NUCB2 (human) mapping to 11p15.1.

PRODUCT

nucleobindin 2 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 nucleobindin 2 shRNA Plasmid (h): sc-62705-SH and nucleobindin 2 shRNA (h) Lentiviral Particles: sc-62705-V as alternate gene silencing products.

For independent verification of nucleobindin 2 (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-62705A, sc-62705B and sc-62705C.

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 2 siRNA (h) is recommended for the inhibition of nucleobindin 2 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

nucleobindin 2 (D-10): sc-376947 is recommended as a control antibody for monitoring of nucleobindin 2 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 2 gene expression knockdown using RT-PCR Primer: nucleobindin 2 (h)-PR: sc-62705-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.