SANTA CRUZ BIOTECHNOLOGY, INC.

Nek11 siRNA (h): sc-78268

BACKGROUND

NIMA (never in mitosis gene A) was originally discovered in *Aspergillus nidulans* as a protein that is necessary for entry into mitosis. Several NIMA-related mammalian proteins have since been identified. Nek11 (NIMA-related kinase 11) is a member of the NIMA subfamily of kinases that functions as a manganese- or magnesium-dependent serine/threonine protein kinase. Kinases of the NIMA subfamily are typically involved in genotoxic stress response and DNA replication. Expressed at low levels in trachea, lung, uterus, appendix and cerebellum, Nek11 localization is cell-cycle regulated; it is found in the nucleus of interphase cells and the polar microtubule of prometaphase and metaphase cells. Nek11 is present in the cell in increasing concentrations from S to G_2/M phase and is believed to play a role in the S phase checkpoint. Two isoforms exist for Nek11, due to alternative splicing events. The long and short isoforms are designated Nek11L and Nek11S, respectively.

REFERENCES

1. Osmani, S.A., et al. 1988. Mitotic induction and maintenance by overexpression of a G_2-specific gene that encodes a potential protein kinase. Cell 53: 237-244.
2. Fry, A.M., et al. 1995. Substrate specificity and cell cycle regulation of the Nek2 protein kinase, a potential human homolog of the mitotic regulator NIMA of *Aspergillus nidulans*. J. Biol. Chem. 270: 12899-12905.
3. Noguchi, K., et al. 2002. Nek11, a new member of the NIMA family of kinases, involved in DNA replication and genotoxic stress responses. J. Biol. Chem. 277: 39655-39665.
4. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 609779. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
5. Belham, C., et al. 2003. A mitotic cascade of NIMA family kinases. Nercc1/Nek9 activates the Nek6 and Nek7 kinases. J. Biol. Chem. 278: 34897-34909.
6. Noguchi, K., et al. 2004. Nucleolar Nek11 is a novel target of Nek2A in G_1/S-arrested cells. J. Biol. Chem. 279: 32716-32727.

CHROMOSOMAL LOCATION

Genetic locus: NEK11 (human) mapping to 3q22.1.

PRODUCT

Nek11 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 Nek11 shRNA Plasmid (h): sc-78268-SH and Nek11 shRNA (h) Lentiviral Particles: sc-78268-V as alternate gene silencing products.

For independent verification of Nek11 (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-78268A, sc-78268B and sc-78268C.

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

Nek11 siRNA (h) is recommended for the inhibition of Nek11 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

Nek11 (36JK): sc-100429 is recommended as a control antibody for monitoring of Nek11 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 Nek11 gene expression knockdown using RT-PCR Primer: Nek11 (h)-PR: sc-78268-PR (20 µl, 463 bp). 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.