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Nek2 siRNA (h): sc-43960

BACKGROUND

NIMA was originally shown in *Aspergillus nidulans* to be necessary for entry into mitosis. NIMA-related mammalian proteins have since been identified as Nek1, Nek2, Nek3 and Nek4 (also designated STK2 or NRK2). High expression of Nek1 is seen in male and female germ cell lines of mouse. Nek2 is the closest known mammalian relative to NIMA. Like NIMA, Nek2 expression peaks at the G₂ to M phase transition. Nek3 is a predominantly cytoplasmic enzyme that was detectable in all organs studied. Levels of Nek3 seem to remain unchanged throughout the cell cycle, but appear to be elevated in G₀-arrested, quiescent fibroblasts. In developing testicular germ cells, differential patterns of expression were seen for Nek1, Nek2 and Nek4, indicating possible overlapping, but non-identical functions.

REFERENCES

- Osmani, S.A., et al. 1988. Mitotic induction and maintenance by overexpression of a G₂-specific gene that encodes a potential protein kinase. *Cell* 53: 237-244.
- Letwin, K., et al. 1992. A mammalian dual specificity protein kinase, Nek1, is related to the NIMA cell cycle regulator and highly expressed in meiotic germ cells. *EMBO J.* 11: 3521-3531.
- Schultz, S.J., et al. 1994. Cell cycle-dependent expression of Nek2, a novel human protein kinase related to the NIMA mitotic regulator of *Aspergillus nidulans*. *Cell Growth Differ.* 5: 625-635.
- Rhee, K., et al. 1997. The NIMA-related kinase 2, Nek2, is expressed in specific stages of the meiotic cell cycle and associates with meiotic chromosomes. *Development* 124: 2167-2177.
- Fry, A.M., et al. 1997. Characterization of mammalian NIMA-related kinases. *Meth. Enzymol.* 283: 270-282.

CHROMOSOMAL LOCATION

Genetic locus: NEK2 (human) mapping to 1q32.3.

PRODUCT

Nek2 siRNA (h) is a target-specific 19-25 nt siRNA 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 Nek2 shRNA Plasmid (h): sc-43960-SH and Nek2 shRNA (h) Lentiviral Particles: sc-43960-V as alternate gene silencing products.

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

Nek2 siRNA (h) is recommended for the inhibition of Nek2 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

Nek2 (D-8): sc-55601 is recommended as a control antibody for monitoring of Nek2 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 Nek2 gene expression knockdown using RT-PCR Primer: Nek2 (h)-PR: sc-43960-PR (20 μl, 537 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- Lai, X.B., et al. 2017. NIMA-related kinase 2 regulates hepatocellular carcinoma cell growth and proliferation. *Oncol. Lett.* 13: 1587-1594.

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.