SANTA CRUZ BIOTECHNOLOGY, INC.

NIFK siRNA (h): sc-72013

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The Power to Question

BACKGROUND

The structural proteins for the complex metalloenzyme nitrogenase include NIFK, NIFD and NIFH. These proteins are all necessary for archaeal and bacterial nitrogen fixation. The NIFK gene encodes the b subunit of the nitrogenase molybdenum-iron (MoFe) tetramer. NIFK localizes to the nucleolus where it interactes with the fork-head associated domain of the proliferation marker protein Ki-67 in a mitosis-specific and phosphorylation-dependent manner. NIFK is widely expressed in adult tissues, suggesting other functions in addition to its interaction with Ki-67, which is only expressed in proliferating cells.

REFERENCES

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2. Ligon, J.M. and Nakas, J.P. 1989. Nucleotide sequence of NIFK and partial sequence of NIFD from Frankia species strain FaC1. Nucleic Acids Res. 16: 11843
3. Li, J.G., et al. 1990. Analysis of Azotobacter vinelandii strains containing defined deletions in the NIFD and NIFK genes. J. Bacteriol. 172: 5884-5891.
4. White, T.C., et al. 1992. Electrophoretic studies on the assembly of the nitrogenase molybdenum-iron protein from the Klebsiella pneumoniae NIFD and NIFK gene products. J. Biol. Chem. 267: 24007-2416.
5. Hirsch, A.M., et al. 1995. Assessing horizontal transfer of NIFHDK genes in eubacteria: nucleotide sequence of NIFK from Frankia strain HFPCcl3. Mol. Biol. Evol. 12: 16-27.
6. Dominic, B., et al. 1998. Cloning and transcriptional analysis of the NIFUHDK genes of Trichodesmium sp. IMS101 reveals stable NIFD, NIFDK and NIFK transcripts. Microbiology 144: 3359-3368.
7. Fani, R., et al. 2000. Molecular evolution of nitrogen fixation: the evolutionary history of the NIFD, NIFK, NIFE, and NIFN genes. J. Mol. Evol. 51: 1-11.
8. Takagi, M., et al. 2001. A novel nucleolar protein, NIFK, interacts with the fork-head associated domain of Ki-67 antigen in mitosis. J. Biol. Chem. 276: 25386-25391.
9. Magnusson, C., et al. 2003. The NIFK gene is widely expressed in mouse tissues and is upregulated in denervated hind limb muscle. Cell Biol. Int. 27: 469-475.

CHROMOSOMAL LOCATION

Genetic locus: MKI67IP (human) mapping to 2q14.3.

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.

PRODUCT

NIFK 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 NIFK shRNA Plasmid (h): sc-72013-SH and NIFK shRNA (h) Lentiviral Particles: sc-72013-V as alternate gene silencing products.

For independent verification of NIFK (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-72013A, sc-72013B and sc-72013C.

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

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

NIFK (18E148): sc-52904 is recommended as a control antibody for monitoring of NIFK 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NIFK gene expression knockdown using RT-PCR Primer: NIFK (h)-PR: sc-72013-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.