

# NRK1 siRNA (m): sc-150069

## BACKGROUND

Nicotinamide adenine dinucleotide (NAD<sup>+</sup>) is an essential cofactor involved in fundamental processes in cell metabolism. NRK1 (nicotinamide riboside kinase 1), also known as ribosylnicotinamide kinase 1, is a 199 amino acid enzyme is involved in the synthesis of NAD<sup>+</sup> through nicotinamide mononucleotide using nicotinamide riboside as the precursor. Nicotinamide riboside has been identified as a nutrient in milk, suggesting that it is a useful compound for elevating the NAD<sup>+</sup> levels in humans. NRK1 also phosphorylates the anti-cancer drugs tiazofurin and 3-deazaguanosine, which converts them into toxic NAD<sup>+</sup> analogs and leads to the inhibition of guanine nucleotide biosynthesis. There are two isoforms of NRK1 that are produced as a result of alternative splicing events.

## REFERENCES

1. Sasiak, K. and Saunders, P.P. 1996. Purification and properties of a human nicotinamide ribonucleoside kinase. *Arch. Biochem. Biophys.* 333: 414-418.
2. Bieganski, P. and Brenner, C. 2004. Discoveries of nicotinamide riboside as a nutrient and conserved NRK genes establish a Preiss-Handler independent route to NAD<sup>+</sup> in fungi and humans. *Cell* 117: 495-502.
3. Online Mendelian Inheritance in Man, OMIM™. 2004. Johns Hopkins University, Baltimore, MD. MIM Number: 608704. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Belenky, P., et al. 2007. Nicotinamide riboside promotes Sir2 silencing and extends lifespan via Nrk and Urh1/Pnp1/Meu1 pathways to NAD<sup>+</sup>. *Cell* 129: 473-484.
5. Ma, B., et al. 2007. Assimilation of NAD<sup>+</sup> precursors in *Candida glabrata*. *Mol. Microbiol.* 66: 14-25.
6. Tempel, W., et al. 2007. Nicotinamide riboside kinase structures reveal new pathways to NAD<sup>+</sup>. *PLoS Biol.* 5: e263.
7. Khan, J.A., et al. 2007. Crystal structure of human nicotinamide riboside kinase. *Structure* 15: 1005-1013.
8. Belenky, P., et al. 2007. NAD<sup>+</sup> metabolism in health and disease. *Trends Biochem. Sci.* 32: 12-19.

## CHROMOSOMAL LOCATION

Genetic locus: Nmrk1 (mouse) mapping to 19 B.

## PRODUCT

NRK1 siRNA (m) is a pool of 2 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 NRK1 shRNA Plasmid (m): sc-150069-SH and NRK1 shRNA (m) Lentiviral Particles: sc-150069-V as alternate gene silencing products.

For independent verification of NRK1 (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-150069A and sc-150069B.

## 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

NRK1 siRNA (m) is recommended for the inhibition of NRK1 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

NRK1 (F-8): sc-398852 is recommended as a control antibody for monitoring of NRK1 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 NRK1 gene expression knockdown using RT-PCR Primer: NRK1 (m)-PR: sc-150069-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](http://www.scbt.com) for detailed protocols and support products.