N-Ras siRNA (h): sc-36004



The Power to Question

BACKGROUND

The mammalian Ras (also designated v-Ha-Ras, Harvey rat sarcoma viral oncogene homolog, HRAS1, K-Ras, N-Ras, RASH1 or c-bas/has) gene family consists of the Harvey and Kirsten Ras genes (c-H-Ras1 and c-K-Ras2), an inactive pseudogene of each (c-H-Ras2 and c-K-Ras1) and the N-Ras gene. The three Ras oncogenes, H-Ras, K-Ras and N-Ras, encode proteins with GTP/GDP binding and GTPase activity. Ras proteins alternate between an inactive form bound to GDP and an active form bound to GTP, activated by a guanine nucleotide-exchange factor (GEF) and inactivated by a GTPaseactivating protein (GAP). Ras nomenclature originates from the characterization of human DNA sequences homologous to cloned DNA fragments containing oncogenic sequences of a type C mammalian retrovirus, the Harvey strain of murine sarcoma virus (HaMSV), derived from the rat. Under normal conditions, Ras family members influence cell growth and differentiation events in a subcellular membrane compartmentalization-based signaling system. Oncogenic Ras can deregulate processes that control both cell proliferation and apoptosis. The Ras superfamily of GTP hydrolysis-coupled signal transduction relay proteins can be subclassified into Ras, Rho, Rab and ARF families.

CHROMOSOMAL LOCATION

Genetic locus: NRAS (human) mapping to 1p13.2.

PRODUCT

N-Ras 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 N-Ras shRNA Plasmid (h): sc-36004-SH and N-Ras shRNA (h) Lentiviral Particles: sc-36004-V as alternate gene silencing products.

For independent verification of N-Ras (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-36004A, sc-36004B and sc-36004C.

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 μ l of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNAse-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

N-Ras siRNA (h) is recommended for the inhibition of N-Ras expression in human cells.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.

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.

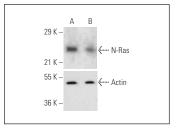
GENE EXPRESSION MONITORING

N-Ras (F155): sc-31 is recommended as a control antibody for monitoring of N-Ras 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 N-Ras gene expression knockdown using RT-PCR Primer: N-Ras (h)-PR: sc-36004-PR (20 μ I, 518 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



N-Ras siRNA (h): sc-36004. Western blot analysis of N-Ras expression in non-transfected control (A) and N-Ras siRNA transfected (B) HeLa cells. Blot probed with N-Ras (F155): sc-31. Actin (I-19): sc-1616 used as spec

SELECT PRODUCT CITATIONS

- 1. Koh, M.S. and Moon, A. 2011. Activation of H-Ras and Rac1 correlates with epidermal growth factor-induced invasion in Hs578T and MDA-MB-231 breast carcinoma cells. Biochem. Biophys. Res. Commun. 406: 25-29.
- Sun, D., et al. 2013. RAS/MEK-independent gene expression reveals BMP2-related malignant phenotypes in the Nf1-deficient MPNST. Mol. Cancer Res. 11: 616-627.
- Ahn, J.H. and Lee, M. 2014. The siRNA-mediated downregulation of N-Ras sensitizes human melanoma cells to apoptosis induced by selective BRAF inhibitors. Mol. Cell. Biochem. 392: 239-247.

RESEARCH USE

For research use only, not for use in diagnostic procedures.