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M-Ras siRNA (h): sc-41857

BACKGROUND

The mammalian c-H-, c-K- and N-Ras proto-oncogenes encode proteins that are ubiquitously expressed in vertebrate cells. c-H- and c-K-Ras are cellular homologs of the v-H- and v-K-Ras sequences originally isolated from the Harvey and Kirsten strains of rat sarcoma virus. Ras p21-encoded proteins bind GDP and GTP with high affinity and possess a low level intrinsic GTPase activity that can be stimulated over 100-fold by interaction with cytosolic GTPase activating protein (GAP), a potential effector for Ras p21 function. Point mutations at amino acids 12, 13, 59 and 61 within domains responsible for GTP binding and hydrolysis, activate Ras proteins to their oncogenic form and block the ability of their GTPase activities to be stimulated by GAP. M-Ras has been identified as a GTPase that shares structural similarities to the Ras family proteins. M-Ras is thought to play a role in reorganization of the Actin cytoskeleton.

REFERENCES

1. Shih, T.Y., et al. 1980. Guanine nucleotide-binding and autophosphorylating activities associated with the p21 Src protein of Harvey murine sarcoma virus. *Nature* 287: 686-691.
2. Ellis, R.W., et al. 1981. The p21 Src genes of Harvey and Kirsten sarcoma viruses originate from divergent members of a family of normal vertebrate genes. *Nature* 292: 506-511.
3. Barbacid, M. 1987. Ras genes. *Annu. Rev. Biochem.* 56: 779-827.
4. Trahey, M. and McCormick, F. 1987. A cytoplasmic protein stimulates normal N-Ras p21 GTPase, but does not affect oncogenic mutants. *Science* 238: 542-545.
5. Calés, C., et al. 1988. The cytoplasmic protein GAP is implicated as the target for regulation by the Ras gene product. *Nature* 332: 548-551.
6. Adari, H., et al. 1988. Guanosine triphosphatase activating protein (GAP) interacts with the p21 Ras effector binding domain. *Science* 240: 518-521.
7. Matsumoto, K., et al. 1997. Novel small GTPase M-Ras participates in reorganization of Actin cytoskeleton. *Oncogene* 15: 2409-2417.

CHROMOSOMAL LOCATION

Genetic locus: MRAS (human) mapping to 3q22.3.

PRODUCT

M-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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see M-Ras shRNA Plasmid (h): sc-41857-SH and M-Ras shRNA (h) Lentiviral Particles: sc-41857-V as alternate gene silencing products.

For independent verification of M-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-41857A, sc-41857B and sc-41857C.

RESEARCH USE

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

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

M-Ras siRNA (h) is recommended for the inhibition of M-Ras 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 μ M in 66 μ l. 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor M-Ras gene expression knockdown using RT-PCR Primer: M-Ras (h)-PR: sc-41857-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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