

E-Ras siRNA (h): sc-60564

BACKGROUND

The Ras-encoded family of proteins bind to GDP and to GTP with high affinity. They possess a low level of intrinsic GTPase activity that can be stimulated more than 100-fold by interaction with cytosolic GTPase activating protein (GAP). Ras family members include H-Ras, K-Ras, N-Ras, M-Ras, R-Ras, E-Ras, Rheb, TC 21, RASL11B and Rad GTPase. H-Ras and K-Ras were first identified as oncogenes of acutely transforming RNA tumor viruses. Subsequently, mutated Ras genes have been found in many human tumors, providing evidence for a common genetic target in cancer. In mammals, a variety of extracellular growth factors that act through protein tyrosine kinase receptors, such as Insulin, platelet-derived growth factor and nerve growth factor, require Ras to exert their effects. Embryonic stem cell-expressed Ras (E-Ras) is a 277 amino acid protein that localizes to the cytoplasmic membrane and shares 43%, 46% and 47% identity with H-Ras, K-Ras and N-Ras, respectively. E-Ras contains five highly conserved domains essential for small G proteins and a CAAX motif.

REFERENCES

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2. Bauer, P.L., Mendeleyeva, J., Kirsten, E., Comstock, J.A., Hakam, A., Buki, K.G. and Kun, E. 2002. Anticancer action of 4-iodo-3-nitrobenzamide in combination with buthionine sulfoximine: inactivation of poly(ADP-ribose) polymerase and tumor glycolysis and the appearance of a poly(ADP-ribose) polymerase protease. *Biochem. Pharmacol.* 63: 455-462.
3. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 300437. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Takahashi, K., Mitsui, K. and Yamanaka, S. 2003. Role of E-Ras in promoting tumour-like properties in mouse embryonic stem cells. *Nature* 423: 541-545.
5. Kameda, T. and Thomson, J.A. 2005. Human E-Ras gene has an upstream premature polyadenylation signal that result in a truncated, noncoding transcript. *Stem Cells* 23: 1535-1540.

CHROMOSOMAL LOCATION

Genetic locus: ERAS (human) mapping to Xp11.23.

PRODUCT

E-Ras siRNA (h) is a pool of 3 target-specific 20-25 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 E-Ras shRNA Plasmid (h): sc-60564-SH and E-Ras shRNA (h) Lentiviral Particles: sc-60564-V as alternate gene silencing products.

For independent verification of E-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-60564A, sc-60564B and sc-60564C.

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

E-Ras siRNA (h) is recommended for the inhibition of E-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 E-Ras gene expression knockdown using RT-PCR Primer: E-Ras (h)-PR: sc-60564-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.