

## ERK 1 siRNA (m2): sc-44206

### BACKGROUND

Mitogen-activated protein kinase (MAPK) signaling pathways involve two closely related MAP kinases, known as extracellular-signal-related kinase 1 (ERK 1, p44) and 2 (ERK 2, p42). Growth factors, steroid hormones, G protein-coupled receptor ligands and neurotransmitters can initiate MAPK signaling pathways. Activation of ERK 1 and ERK 2 requires phosphorylation by upstream kinases such as MAP kinasekinase (MEK), MEK kinase and Raf-1. ERK 1 and ERK 2 phosphorylation can occur at specific tyrosine and threonine sites mapping within consensus motifs that include the threonine-glutamate-tyrosine motif. ERK activation leads to dimerization with other ERKs and subsequent localization to the nucleus. Active ERK dimers phosphorylate serine and threonine residues on nuclear proteins and influence a host of responses that include proliferation, differentiation, transcription regulation and development. The human ERK 1 gene maps to chromosome 16p11.2 and encodes a 379 amino acid protein that shares 83% sequence identity to ERK 2.

### REFERENCES

1. Boulton, T.G., et al. 1991. ERKs: a family of protein-serine/threonine kinases that are activated and tyrosine phosphorylated in response to Insulin and NGF. *Cell* 65: 663-675.
2. Crews, C.M., et al. 1992. The primary structure of MEK, a protein kinase that phosphorylates the ERK gene product. *Science* 258: 478-480.
3. Haycock, J.W., et al. 1992. ERK 1 and ERK 2, two microtubule-associated protein 2 kinases, mediate the phosphorylation of tyrosine hydroxylase at Serine 31 *in situ*. *Proc. Natl. Acad. Sci. USA* 89: 2365-2369.
4. Charest, D.L., et al. 1993. Molecular cloning, expression, and characterization of the human mitogen-activated protein kinase p44 ERK 1. *Mol. Cell. Biol.* 13: 4679-4690.
5. Khokhlatchev, A.V., et al. 1998. Phosphorylation of the MAP kinase ERK 2 promotes its homodimerization and nuclear translocation. *Cell* 93: 605-615.
6. Pages, G., et al. 2000. Signaling angiogenesis via p42/p44 MAP kinase cascade. *Ann. N.Y. Acad. Sci.* 902: 187-200.
7. Gutkind, J.S. 2000. Regulation of mitogen-activated protein kinase signaling networks by G protein-coupled receptors. *Sci. STKE* 2000: re1.

### CHROMOSOMAL LOCATION

Genetic locus: Mapk3 (mouse) mapping to 7 F3.

### PRODUCT

ERK 1 siRNA (m2) 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ERK 1 shRNA Plasmid (m2): sc-44206-SH and ERK 1 shRNA (m2) Lentiviral Particles: sc-44206-V as alternate gene silencing products.

For independent verification of ERK 1 (m2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44206A and sc-44206B.

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

### APPLICATIONS

ERK 1 siRNA (m2) is recommended for the inhibition of ERK 1 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  $\mu$ M in 66  $\mu$ 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.

### GENE EXPRESSION MONITORING

ERK 1 (G-8): sc-271269 is recommended as a control antibody for monitoring of ERK 1 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 ERK 1 gene expression knockdown using RT-PCR Primer: ERK 1 (m2)-PR: sc-44206-PR (20  $\mu$ l, 452 bp). 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.