

MLK3 siRNA (m): sc-35946

BACKGROUND

As a result of the binding of growth factors to their membrane receptors, cytoplasmic proteins containing Src homology 2 (SH2) domains associate with specific phosphotyrosine residues within the activated receptors and function as signaling intermediates. The action of such SH2 domain proteins frequently involves the activation of a second group of signaling intermediates characterized by SH3 domains. These latter proteins function through binding proline-rich sequences in target proteins. A novel human non-receptor protein kinase, designated either MLK3 or PTK1, is 847 amino acids in length and contains an SH3 domain in the absence of an SH2 domain. In addition, MLK3 is characterized by a leucine zipper basic region (a motif commonly associated with transcription factors) and has a long carboxy-terminal tail which exhibits proline-rich motifs similar to known SH3 binding sites. MLK3 is expressed widely and is related to the previously described MLK1 and MLK2 kinases.

REFERENCES

- Schlessinger, J., et al. 1992. Growth factor signaling by receptor tyrosine kinases. *Neuron* 9: 383-391.
- Mayer, B., et al. 1993. Signalling through SH2 and SH3 domains. *Trends Cell Biol.* 3: 8-13.
- Pawson, T., et al. 1993. Proteins with SH2 and SH3 domains couple receptor tyrosine kinases to intracellular signalling pathways. *Philos. Trans. R. Soc. Lond. B, Biol. Sci.* 340: 279-285.
- Dorow, D.S., et al. 1993. Identification of a new family of human epithelial protein kinases containing two leucine/isoleucine-zipper domains. *Eur. J. Biochem.* 213: 701-710.
- Ing, Y.L., et al. 1994. MLK3: identification of a widely-expressed protein kinase bearing an SH3 domain and a leucine zipper-basic region domain. *Oncogene* 9: 1745-1750.
- Ezoe, K., et al. 1994. PTK1, a novel protein kinase required for proliferation of human melanocytes. *Oncogene* 9: 935-938.

CHROMOSOMAL LOCATION

Genetic locus: Map3k11 (mouse) mapping to 19 A.

PRODUCT

MLK3 siRNA (m) 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 MLK3 shRNA Plasmid (m): sc-35946-SH and MLK3 shRNA (m) Lentiviral Particles: sc-35946-V as alternate gene silencing products.

For independent verification of MLK3 (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-35946A, sc-35946B and sc-35946C.

PROTOCOLS

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

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

MLK3 siRNA (m) is recommended for the inhibition of MLK3 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 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.

GENE EXPRESSION MONITORING

MLK3 (D-11): sc-166639 is recommended as a control antibody for monitoring of MLK3 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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 MLK3 gene expression knockdown using RT-PCR Primer: MLK3 (m)-PR: sc-35946-PR (20 μ l). 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.