

MLK4 siRNA (h): sc-61061

BACKGROUND

Mixed lineage kinases are a family of protein kinases sharing two leucine zipper-like motifs, which are known to mediate protein dimerization and a kinase domain whose primary structure is similar to both the tyrosine-specific and the serine/threonine-specific kinase classes. Members of the mixed-lineage kinase (MLK) family include MLK1, MLK2, MLK3, MLK4, MELK, LZK and dual leucine zipper kinase, also designated DLK. MLKs are expressed in neuronal cells where they are likely to interact between Rac1/Cdc42, MKK4 and MKK7 in death signaling.

REFERENCES

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2. Nagata, K., et al. 1998. The MAP kinase kinase kinase MLK2 co-localizes with activated JNK along microtubules and associates with kinesin superfamily motor KIF3. *EMBO J.* 17: 149-158.
3. Leung, I.W. and Lassam, N. 1999. Dimerization via tandem leucine zippers is essential for the activation of the mitogen-activated protein kinase kinase kinase, MLK-3. *J. Biol. Chem.* 273: 32408-32415.
4. Xu, Z., et al. 2001. The MLK family mediates c-Jun N-terminal kinase activation in neuronal apoptosis. *Mol. Cell. Biol.* 21: 4713-4724.
5. Gallo, K.A. and Johnson, G.L. 2002. Mixed-lineage kinase control of JNK and p38 MAPK pathways. *Mol. Cell. Biol.* 3: 663-672.
6. Falsig, J., et al. 2004. Specific modulation of astrocyte inflammation by inhibition of mixed lineage kinases with CEP-1347. *J. Immunol.* 173: 2762-2770.
7. Soung, Y.H., et al. 2006. Kinase domain mutation of MLK4 gene is uncommon in gastric and hepatocellular carcinomas. *Dig. Liver Dis.* 38: 283.

CHROMOSOMAL LOCATION

Genetic locus: MAP3K21 (human) mapping to 1q42.2.

PRODUCT

MLK4 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 MLK4 shRNA Plasmid (h): sc-61061-SH and MLK4 shRNA (h) Lentiviral Particles: sc-61061-V as alternate gene silencing products.

For independent verification of MLK4 (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-61061A, sc-61061B and sc-61061C.

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

MLK4 siRNA (h) is recommended for the inhibition of MLK4 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.

GENE EXPRESSION MONITORING

MLK4 (E-11): sc-398697 is recommended as a control antibody for monitoring of MLK4 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 MLK4 gene expression knockdown using RT-PCR Primer: MLK4 (h)-PR: sc-61061-PR (20 μ l, 555 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.