

MP1 siRNA (h): sc-40746

BACKGROUND

MP1 (MEK partner 1) functions as a scaffolding protein in the mitogen activated protein (MAP) kinase signaling pathway. Growth factor induced MAP kinase activation is selectively mediated by the extracellular signal-regulated kinase (ERK) cascade. This pathway is dependent on the phosphorylation of MEK-1 and its subsequent activation of ERK 1. MP1 binds to the proline-rich domain of MEK-1 and thereby potentiates the phosphorylation of MEK-1 by the activating MEK kinase B-Raf. MP1 is also able to enhance the kinase activity of MEK-1 and facilitate the phosphorylation of ERK 1. *In vivo* studies indicate that MP1 preferentially associates with MEK-1 and ERK 1, but not with MEK-2 or ERK 2, suggesting that MP1 and other scaffolding proteins contribute to the specificity of the kinase substrates within the MAPK pathways.

REFERENCES

1. Elion, E.A. 1998. Routing MAP kinase cascades. *Science* 281: 1625-1626.
2. Schaeffer, H.J., et al. 1998. MP1: a MEK binding partner that enhances enzymatic activation of the MAP kinase cascade. *Science* 281: 1668-1671.
3. Whitmarsh, A.J., et al. 1998. A mammalian scaffold complex that selectively mediates MAP kinase activation. *Science* 281: 1671-1674.
4. Garrington, T.P. and Johnson, G.L. 1999. Organization and regulation of mitogen-activated protein kinase signaling pathways. *Curr. Opin. Cell Biol.* 11: 211-218.
5. Schaeffer, H.J. and Weber, M.J. 1999. Mitogen-activated protein kinases: specific messages from ubiquitous messengers. *Mol. Cell. Biol.* 19: 2435-2444.
6. Sharma, C., et al. 2005. MEK partner 1 (MP1): regulation of oligomerization in MAP kinase signaling. *J. Cell. Biochem.* 94: 708-719.
7. Pullikuth, A., et al. 2005. The MEK1 scaffolding protein MP1 regulates cell spreading by integrating PAK1 and Rho signals. *Mol. Cell. Biol.* 25: 5119-5133.
8. Mouchel-Vielh, E., et al. 2008. Involvement of the MP1 scaffold protein in ERK signaling regulation during *Drosophila* wing development. *Genes Cells* 13: 1099-1111.

CHROMOSOMAL LOCATION

Genetic locus: LAMTOR3 (human) mapping to 4q23.

PRODUCT

MP1 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 MP1 shRNA Plasmid (h): sc-40746-SH and MP1 shRNA (h) Lentiviral Particles: sc-40746-V as alternate gene silencing products.

For independent verification of MP1 (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-40746A, sc-40746B and sc-40746C.

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

MP1 siRNA (h) is recommended for the inhibition of MP1 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

MP1 (H-6): sc-376783 is recommended as a control antibody for monitoring of MP1 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 MP1 gene expression knockdown using RT-PCR Primer: MP1 (h)-PR: sc-40746-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.

PROTOCOLS

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