

Lamellipodin siRNA (h): sc-62539

BACKGROUND

Lamellipodin, also called Ras-associated and Pleckstrin homology domains-containing protein 1 (RAPH1), is a 1,302 amino acid member of the MRL family. The peripheral membrane protein mediates localized membrane signals and co-localizes at the tips of filopodia and lamellipodia with ENAH/VASP. Aside from the main isoform, nine additional isoforms have been identified for Lamellipodin (RMO1, RMO1a, RMO1b, RMO1c, RMO1ab, RMO1ac, RMO1bc, RMO1abc and RMO1-RAPH1). RMO1-RAPH1 is expressed in a wide variety of tissues, most highly in brain, heart, ovary and developing embryo. RMO1 is also widely expressed, with highest amounts in liver. Lamellipodin is down-regulated in breast and ovarian cancers and shows reduced expression in metastatic osteosarcomas in comparison to primary osteosarcoma tumors.

REFERENCES

1. Hadano, S., et al. 2001. A gene encoding a putative GTPase regulator is mutated in familial amyotrophic lateral sclerosis 2. *Nat. Genet.* 29: 166-173.
2. Krause, M., et al. 2004. Lamellipodin, an ENA/VASP ligand, is implicated in the regulation of lamellipodial dynamics. *Dev. Cell* 7: 571-583.
3. Eppert, K., et al. 2005. Altered expression and deletion of RMO1 in osteosarcoma. *Int. J. Cancer* 114: 738-746.
4. Adler, C.E., et al. 2006. UNC-6/netrin induces neuronal asymmetry and defines the site of axon formation. *Nat. Neurosci.* 9: 511-518.
5. Quinn, C.C., et al. 2006. UNC-6/netrin and SLT-1/Slit guidance cues orient axon outgrowth mediated by MIG-10/RIAM/Lamellipodin. *Curr. Biol.* 16: 845-853.
6. Chang, C., et al. 2006. MIG-10/Lamellipodin and AGE-1/PI 3-K promote axon guidance and outgrowth in response to Slit and netrin. *Curr. Biol.* 16: 854-862.
7. Sjöblom, T., et al. 2006. The consensus coding sequences of human breast and colorectal cancers. *Science* 314: 268-274.

CHROMOSOMAL LOCATION

Genetic locus: RAPH1 (human) mapping to 2q33.2.

PRODUCT

Lamellipodin 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 Lamellipodin shRNA Plasmid (h): sc-62539-SH and Lamellipodin shRNA (h) Lentiviral Particles: sc-62539-V as alternate gene silencing products.

For independent verification of Lamellipodin (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-62539A, sc-62539B and sc-62539C.

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

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

Lamellipodin (H-5): sc-390050 is recommended as a control antibody for monitoring of Lamellipodin 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 Lamellipodin gene expression knockdown using RT-PCR Primer: Lamellipodin (h)-PR: sc-62539-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.