

Formin 1 siRNA (h): sc-89914

BACKGROUND

The temporal genetic hierarchy influencing normal limb development can deregulate and mediate mammalian developmental syndromes. In mice, the limb deformity (Id) locus influences normal limb development and gives rise to alternative mRNAs that can translate into a family of proteins known as formins. Formins play a crucial role in cytoskeletal reorganization by influencing Actin filament assembly. Formins co-localize with the Actin cytoskeleton and can translocate into the cell cytosol and into the nucleus in an HGF-dependent manner. Vertebrate nuclear formins can control polarizing activity in limb buds through establishment of a Sonic hedgehog/FGF-4 feedback loop. Deficiency mutations at the mammalian Id locus lead to profound developmental defects in limb and kidney formation. The human Formin 1 and 2 genes map to chromosome 15q13.3 and 1q43, respectively.

REFERENCES

1. Maas, R.L., Jepeal, L.I., Elfering, S.L., Holcombe, R.F., Morton, C.C., Eddy, R.L., Byers, M.G., Shows, T.B. and Leder, P. 1991. A human gene homologous to the formin gene residing at the murine limb deformity locus: chromosomal location and RFLPs. *Am. J. Hum. Genet.* 48: 687-695.
2. Wynshaw-Boris, A., Ryan, G., Deng, C.X., Chan, D.C., Jackson-Grusby, L., Larson, D., Dunmore, J.H. and Leder, P. 1997. The role of a single formin isoform in the limb and renal phenotypes of limb deformity. *Mol. Med.* 3: 372-384.
3. Zeller, R., Haramis, A.G., Zuniga, A., McGuigan, C., Dono, R., Davidson, G., Chabanis, S. and Gibson, T. 1999. Formin defines a large family of morphoregulatory genes and functions in establishment of the polarising region. *Cell Tissue Res.* 296: 85-93.
4. Tanaka, K. 2000. Formin family proteins in cytoskeletal control. *Biochem. Biophys. Res. Commun.* 267: 479-481.
5. O'Rourke, D.A., Liu, Z.X., Sellin, L., Spokes, K., Zeller, R. and Cantley, L.G. 2000. Hepatocyte growth factor induces MAPK-dependent Formin IV translocation in renal epithelial cells. *J. Am. Soc. Nephrol.* 11: 2212-2221.

CHROMOSOMAL LOCATION

Genetic locus: FMN1 (human) mapping to 15q13.3.

PRODUCT

Formin 1 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 Formin 1 shRNA Plasmid (h): sc-89914-SH and Formin 1 shRNA (h) Lentiviral Particles: sc-89914-V as alternate gene silencing products.

For independent verification of Formin 1 (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-89914A, sc-89914B and sc-89914C.

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

Formin 1 siRNA (h) is recommended for the inhibition of Formin 1 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

Formin 1 (H-5): sc-515698 is recommended as a control antibody for monitoring of Formin 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 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 Formin 1 gene expression knockdown using RT-PCR Primer: Formin 1 (h)-PR: sc-89914-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.