



Lunatic Fringe siRNA (m): sc-39491

BACKGROUND

Three mammalian fringe family members, namely Manic Fringe, Radical Fringe and Lunatic Fringe, have been identified as proteins related to *Drosophila* fringe, a protein involved in development. Fringe proteins act upstream of the Notch signaling pathway and are involved in boundary determination during segmentation. Each mammalian fringe displays different patterns of expression, though all are expressed in the mouse embryo as well as in many adult tissues. Lunatic Fringe, also known as LFNG or SCD03, is a 379 amino acid single-pass type II membrane protein that localizes to the membrane of the Golgi apparatus. Functioning as a glycosyltransferase, Lunatic Fringe acts a critical mediator of somite patterning and segmentation and plays a fundamental role in initiating the elongation of O-linked fucose residues that are attached to Notch molecules. Defects in the gene encoding Lunatic Fringe are the cause of autosomal recessive spondylocostal dysostosis 3 (SCDO3), a disorder that arises during embryonic development and is characterized by rib anomalies and multiple vertebrate segmentation. Three isoforms of Lunatic Fringe are expressed due to alternative splicing events.

REFERENCES

1. Johnston, S.H., et al. 1997. A family of mammalian Fringe genes implicated in boundary determination and the Notch pathway. *Development* 124: 2245-2254.
2. May, W.A., et al. 1997. EWS/FLI1-induced manic fringe renders NIH 3T3 cells tumorigenic. *Nat. Genet.* 17: 495-497.
3. Laufer, E., et al. 1997. Expression of Radical fringe in limb-bud ectoderm regulates apical ectodermal ridge formation. *Nature* 386: 366-373.
4. Thelu, J., et al. 1998. Differential expression pattern of the three fringe genes is associated with epidermal differentiation. *J. Invest. Dermatol.* 111: 903-906.
5. Evrard, Y.A., et al. 1998. Lunatic fringe is an essential mediator of somite segmentation and patterning. *Nature* 394: 377-381.
6. McGrew, M.J., et al. 1998. The lunatic fringe gene is a target of the molecular clock linked to somite segmentation in avian embryos. *Curr. Biol.* 8: 979-982.

CHROMOSOMAL LOCATION

Genetic locus: *Lfng* (mouse) mapping to 5 G2.

PRODUCT

Lunatic Fringe 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 Lunatic Fringe shRNA Plasmid (m): sc-39491-SH and Lunatic Fringe shRNA (m) Lentiviral Particles: sc-39491-V as alternate gene silencing products.

For independent verification of Lunatic Fringe (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-39491A, sc-39491B and sc-39491C.

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

Lunatic Fringe siRNA (m) is recommended for the inhibition of Lunatic Fringe 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

Lunatic Fringe (TT-07): sc-100756 is recommended as a control antibody for monitoring of Lunatic Fringe 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 Lunatic Fringe gene expression knockdown using RT-PCR Primer: Lunatic Fringe (m)-PR: sc-39491-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.