

Filamin 3 siRNA (h): sc-60641

BACKGROUND

Filamins are Actin-binding proteins which contain an N-terminal Actin-binding domain, a membrane glycoprotein domain and a C-terminal self-association domain. Filamins help reshape the cytoskeleton by forming flexible cross-links between two Actin filaments, which maintain membrane integrity during force application. Filamins also participate in signal transduction pathways associated with cell motility, adhesion, differentiation and survival, and force transduction. The Filamin family is comprised of Filamin 1, Filamin 2 and Filamin 3. Filamin 3, also designated Filamin B and β -Filamin, is a form of Filamin that plays a role in endochondral ossification, vertebral segmentation and joint formation. The interaction of Filamin 3 with Filamin 1 may allow neuroblast migration into the cortical plate from the ventricular zone. Mutations in the gene that encodes for Filamin 3, FLNB, are associated with five human skeletal disorders, specifically, autosomal dominant Larsen syndrome, spondylarctoparsal syndrome, type I atelosteogenesis, type III atelosteogenesis and boomerang dysplasia as well as the neurologic disorder periventricular heterotopia.

REFERENCES

1. Takafuta, T., et al. 1998. Human β -Filamin is a new protein that interacts with the cytoplasmic tail of glycoprotein Ib α . *J. Biol. Chem.* 273: 17531-17538.
2. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603381. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Krakow, D., et al. 2004. Mutations in the gene encoding Filamin B disrupt vertebral segmentation, joint formation and skeletogenesis. *Nat. Genet.* 36: 405-410.
4. Bicknell, L.S., et al. 2005. Mutations in FLNB cause boomerang dysplasia. *J. Med. Genet.* 42: E43.
5. Ohashi, K., et al. 2005. Chicken gizzard filamin, retina filamin and cgABP260 are respectively, smooth muscle-, non-muscle- and pan-muscle-type isoforms: distribution and localization in muscles. *Cell Motil. Cytoskeleton* 61: 214-225.
6. Pudas, R., et al. 2005. Structural basis for vertebrate filamin dimerization. *Structure* 13: 111-119.

CHROMOSOMAL LOCATION

Genetic locus: FLNB (human) mapping to 3p14.3.

PRODUCT

Filamin 3 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 Filamin 3 shRNA Plasmid (h): sc-60641-SH and Filamin 3 shRNA (h) Lentiviral Particles: sc-60641-V as alternate gene silencing products.

For independent verification of Filamin 3 (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-60641A, sc-60641B and sc-60641C.

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

Filamin 3 siRNA (h) is recommended for the inhibition of Filamin 3 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

Filamin 3 (F-8): sc-376241 is recommended as a control antibody for monitoring of Filamin 3 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 Filamin 3 gene expression knockdown using RT-PCR Primer: Filamin 3 (h)-PR: sc-60641-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.