SANTA CRUZ BIOTECHNOLOGY, INC.

Myosin IIIa siRNA (h): sc-90539

BACKGROUND

Myosins are highly conserved, ubiquitously expressed proteins that interact with Actin to generate the force for cellular movements. The human genome encodes over 40 different Myosin genes which are divided into distinct classes, the most notable of which are the conventional Myosins (class II) and the unconventional Myosins (classes I and III through XV). Myosin IIIA, also known as MYO3A or DFNB30, is a 1,616 amino acid class III Myosin that contains one protein kinase domain, one myosin head-like domain and three IQ domains. Highly expressed in pancreas, cochlea and retinal pigment cells, Myosin IIIA is thought to function as an Actin-based motor that may play a role in both vision and hearing. Additionally, via its protein kinase domain, Myosin IIIA is able to catalyze the ATP-dependent phosphorylation of a variety of target proteins. Defects in the gene encoding Myosin IIIA are the cause of non-syndromic sensorineural deafness autosomal recessive type 30 (DFNB30), a form of hearing loss that is caused by damage to neural receptors in the brain or inner ear.

REFERENCES

1. Dose, A.C. and Burnside, B. 2000. Cloning and chromosomal localization of a human class III Myosin. Genomics 67: 333-342.
2. Walsh, T., et al. 2002. From flies' eyes to our ears: mutations in a human class III Myosin cause progressive nonsyndromic hearing loss DFNB30. Proc. Natl. Acad. Sci. USA 99: 7518-7523.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 606808. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Komaba, S., et al. 2003. Determination of human Myosin III as a motor protein having a protein kinase activity. J. Biol. Chem. 278: 21352-21360.
5. Les Erickson, F., et al. 2003. Localization of a class III Myosin to filopodia tips in transfected HeLa cells requires an Actin-binding site in its tail domain. Mol. Biol. Cell 14: 4173-4180.
6. Kambara, T., et al. 2006. Human myosin III is a motor having an extremely high affinity for actin. J. Biol. Chem. 281: 37291-37301.
7. Schneider, M.E., et al. 2006. A new compartment at stereocilia tips defined by spatial and temporal patterns of Myosin IIIa expression. J. Neurosci. 26: 10243-10252.

CHROMOSOMAL LOCATION

Genetic locus: MYO3A (human) mapping to 10p12.1.

PRODUCT

Myosin IIIa 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Myosin IIIa shRNA Plasmid (h): sc-90539-SH and Myosin IIIa shRNA (h) Lentiviral Particles: sc-90539-V as alternate gene silencing products.

For independent verification of Myosin IIIa (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-90539A, sc-90539B and sc-90539C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Myosin IIIa siRNA (h) is recommended for the inhibition of Myosin IIIa 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 µM in 66 µl. 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

Myosin IIIa (RR-8): sc-100953 is recommended as a control antibody for monitoring of Myosin IIIa 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Myosin IIIa gene expression knockdown using RT-PCR Primer: Myosin IIIa (h)-PR: sc-90539-PR (20 µl, 401 bp). 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.