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Myosin Ie siRNA (h): sc-44613

BACKGROUND

Actin is a highly conserved protein that is expressed in all eukaryotic cells. Actin filaments can form both stable and labile structures and are crucial components of microvilli and the contractile apparatus of muscle cells. Troponin facilitates interaction between Actin and Myosin by binding to Ca^{2+} . Troponin is made up of at least two subunits, which are divergent in cardiac muscle, fast skeletal muscle and slow skeletal muscle. Myosin is a hexamer of two heavy chains (MHC) and four light chains (MLC) that interacts with Actin to generate the force for diverse cellular movements, including cytoskeleton, phagocytosis and muscle contraction. Myosin Ie (MYO1E) is also designated Myosin Ic. Myosin Ie contains 1,109 amino acids.

REFERENCES

- Whitmer, J.D., et al. 1996. Chromosomal location of three unconventional myosin heavy chain genes in the mouse. *Genomics* 38: 235-237.
- Dumont, R.A., et al. 2002. Myosin I isozymes in neonatal rodent auditory and vestibular epithelia. *J. Assoc. Res. Otolaryngol.* 3: 375-389.
- El Mezgueldi, M., et al. 2002. The kinetic mechanism of Myo1e (human Myosin Ic). *J. Biol. Chem.* 277: 21514-21521.
- Lindvall, J.M., et al. 2005. Differential expression and molecular characterization of Lmo7, Myo1e, Sash1 and Mcoln2 genes in Btk-defective B cells. *Cell. Immunol.* 235: 46-55.
- SWISS-PROT/TrEMBL (Q12965). World Wide Web URL: <http://www.xpasy.ch/sprot/sprot-top.html>

CHROMOSOMAL LOCATION

Genetic locus: MYO1E (human) mapping to 15q22.2.

PRODUCT

Myosin Ie 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 Myosin Ie shRNA Plasmid (h): sc-44613-SH and Myosin Ie shRNA (h) Lentiviral Particles: sc-44613-V as alternate gene silencing products.

For independent verification of Myosin Ie (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-44613A, sc-44613B and sc-44613C.

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

Myosin Ie siRNA (h) is recommended for the inhibition of Myosin Ie 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

Myosin Ie (7A5): sc-293354 is recommended as a control antibody for monitoring of Myosin Ie 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 Myosin Ie gene expression knockdown using RT-PCR Primer: Myosin Ie (h)-PR: sc-44613-PR (20 μ l, 392 bp). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ 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.