SANTA CRUZ BIOTECHNOLOGY, INC.

MYH1 siRNA (m): sc-35930

BACKGROUND

Myosin is a highly conserved, ubiquitously expressed protein that interacts with Actin to generate the force for cellular movements. Conventional myosins are hexameric proteins consisting of two heavy chain subunits, a pair of non-phosphorylatable light chain subunits and a pair of phosphorylatable light chain subunits. Three general classes of myosin have been cloned: smooth muscle myosins, striated muscle myosins and non-muscle myosins. Contractile activity in smooth muscle is regulated by the calcium/calmodulin-dependent phosphorylation of Myosin light chain (MLC) by Myosin light chain kinase. Myosin heavy chains (MHCs) contain Actin-activated ATPase activity which generates the motor function of Myosin. MHCs, which were initially isolated from a human fetal skeletal muscle, are the major determinant in the speed of contraction of skeletal muscle, and various isoforms are differentially expressed depending on the functional activity of the muscle. The MYH1 (skeletal Myosin heavy chain 1) locus is part of a family of six genes clustered on a single chromosome, 17p in human and 11 in mouse, which are abundantly expressed in skeletal muscle.

REFERENCES

1. Nagai, R., et al. 1989. Vertebrate smooth muscle Myosin heavy chains (MHCs) exist as two isoforms with molecular masses of 204 and 200 kDa (MHC204 and MHC200) that are generated from a single gene by alternative splicing of mRNA. J. Biol. Chem. 264: 9734-9737.
2. Karsch-Mizrachi, I., et al. 1990. Generation of a full-length human perinatal Myosin heavy-chain-encoding cDNA. Gene 89: 289-294.
3. Bober, E., et al. 1990. Identification of three developmentally controlled isoforms of human Myosin heavy chains. Eur. J. Biochem. 189: 55-65.
4. Yoon, S.J., et al. 1992. Organization of the human skeletal Myosin heavy chain gene cluster. Proc. Natl. Acad. Sci. USA 89: 12078-12082.
5. Cheney, R.E., et al. 1993. Phylogenetic analysis of the Myosin superfamily. Cell Motil. Cytoskelet. 24: 215-223.
6. Jullian, E.H., et al. 1995. Characterization of a human perinatal Myosin heavy-chain transcript. Eur. J. Biochem. 230: 1001-1006.
7. Owens, G.K. 1995. Regulation of differentiation of vascular smooth muscle cells. Physiol. Rev. 75: 487-517.

CHROMOSOMAL LOCATION

Genetic locus: Myh1 (mouse) mapping to 11 B3.

PRODUCT

MYH1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see MYH1 shRNA Plasmid (m): sc-35930-SH and MYH1 shRNA (m) Lentiviral Particles: sc-35930-V as alternate gene silencing products.

For independent verification of MYH1 (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-35930A, sc-35930B and sc-35930C.

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

MYH1 siRNA (m) is recommended for the inhibition of MYH1 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 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

MYH1/2/3 (N3.36): sc-53092 is recommended as a control antibody for monitoring of MYH1 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 MYH1 gene expression knockdown using RT-PCR Primer: MYH1 (m)-PR: sc-35930-PR (20 µl, 566 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.