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SANTA CRUZ BIOTECHNOLOGY, INC.

Myotilin siRNA (m): sc-43409

BACKGROUND

Myotilin, a sarcomeric protein that is encoded by the gene mapping to human chromosome 5q31.2, binds to α-actinin and is localized in the Z-line of myofibrils. Myotilin is expressed in skeletal and cardiac muscle, and it co-localizes with α-actinin in the sarcomeric I-bands where it directly interacts with α-actinin. Defects in the myotilin gene are reported to cause a form of autosomal dominant limb-girdle muscular dystrophy (LGMD). Symptoms of adult onset LGMD are progressive weakness of the hip and shoulder girdles as well as a distinctive dysarthric pattern of speech. The muscle of affected individuals with LGMD shows degeneration of myofibers, variations in fiber size, fiber splitting, centrally located myonuclei and an enhanced number of autophagic vesicles.

REFERENCES

1. Speer, M.C., et al. 1995. Evidence for locus heterogeneity in autosomal dominant limb-girdle muscular dystrophy. Am. J. Hum. Genet. 57: 1371-1376.
2. Minetti, C., et al. 1998. Mutations in the caveolin-3 gene cause autosomal dominant limb-girdle muscular dystrophy. Nat. Genet. 18: 365-368.
3. Salmikangas, P., et al. 1999. Myotilin, a novel sarcomeric protein with two Ig-like domains, is encoded by a candidate gene for limb-girdle muscular dystrophy. Hum. Mol. Genet. 8: 1329-1336.
4. van der Ven, P.F., et al. 2000. Indications for a novel muscular dystrophy pathway. γ-filamin, the muscle-specific filamin isoform, interacts with Myotilin. J. Cell Biol. 151: 235-248.
5. Hauser, M.A., et al. 2000. Myotilin is mutated in limb girdle muscular dystrophy 1A. Hum. Mol. Genet. 9: 2141-2147.

CHROMOSOMAL LOCATION

Genetic locus: Myot (mouse) mapping to 18 B3.

PRODUCT

Myotilin 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 Myotilin shRNA Plasmid (m): sc-43409-SH and Myotilin shRNA (m) Lentiviral Particles: sc-43409-V as alternate gene silencing products.

For independent verification of Myotilin (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-43409A, sc-43409B and sc-43409C.

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.

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

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

Myotilin (E-10): sc-393957 is recommended as a control antibody for monitoring of Myotilin 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 Myotilin gene expression knockdown using RT-PCR Primer: Myotilin (m)-PR: sc-43409-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.