SANTA CRUZ BIOTECHNOLOGY, INC.

MYL5 siRNA (h): sc-61126

BACKGROUND

Myosin interacts with actin to generate the force for diverse cellular movements, including cytokinesis, phagocytosis and muscle contraction. Myosin is a hexamer of two heavy chains (MHC) and four light chains (MLC), two of which are nonphosphorylatable alkali light chains and the other two are phosphorylatable regulatory light chains. Myosin regulatory light chain 5, also known as Myosin LC2, is encoded by the MYL5 gene and expressed in fetal muscle as well as adult retina, cerebellum, and basal ganglia. Removal of light chains from myosin reduces the velocity of Actin filaments. Reconstitution of myosin with regulatory light chain 5 or alkali light chain increases filament velocity to intermediate rates, and readdition of both classes of light chains fully restores the original sliding velocity.

REFERENCES

1. Kuo, T.H. and Banerjee, S.K. 1983. Effects of removal of light chain 2 on the ATPase activities of cardiac myosin from normal and thyrotoxic rabbits. Biochim. Biophys. Acta 707: 199-205.
2. Lowey, S., et al. 1993. Skeletal muscle myosin light chains are essential for physiological speeds of shortening. Nature 365: 454-456.
3. Collins, C., et al. 1993. The genomic organization of a novel regulatory myosin light chain gene (MYL5) that maps to chromosome 4p16.3 and shows different patterns of expression between primates. Hum. Mol. Genet. 1: 727-733.
4. Roulet, A., et al. 1993. The proteolytic susceptibility of specific sites in myosin light chains is modulated by the filament conformation. Eur. J. Biochem. 216: 89-101.
5. Holt, J.C., et al. 1995. Human cardiac myosin light chains: sequence comparisons between Myosin LC1 and LC2 from normal and idiopathic dilated cardiomyopathic hearts. Mol. Cell. Biochem. 145: 89-96.
6. Stepkowski, D., et al. 1995. Significance of the N-terminal fragment of myosin regulatory light chain for myosin-Actin interaction. Biochem. Mol. Biol. Int. 35: 677-684.
7. Khalina, IaN., et al. 2002. Changes in composition of cardiac myosin light chains in dilated cardiomyopathy: effect on functional properties. Biofizika 47: 361-366.

CHROMOSOMAL LOCATION

Genetic locus: MYL5 (human) mapping to 4p16.3.

PRODUCT

MYL5 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 MYL5 shRNA Plasmid (h): sc-61126-SH and MYL5 shRNA (h) Lentiviral Particles: sc-61126-V as alternate gene silencing products.

For independent verification of MYL5 (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-61126A, sc-61126B and sc-61126C.

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

MYL5 siRNA (h) is recommended for the inhibition of MYL5 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

MYL5 (SB-19): sc-100952 is recommended as a control antibody for monitoring of MYL5 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 MYL5 gene expression knockdown using RT-PCR Primer: MYL5 (h)-PR: sc-61126-PR (20 µl). 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.