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SANTA CRUZ BIOTECHNOLOGY, INC.

Myosin Ib shRNA (m) Lentiviral Particles: sc-44601-V



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 Ca²⁺. 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 cytokinesis, phagocytosis and muscle contraction. Myosin Ib (MY01B), also designated Myosin I α or MYH-1c, is a motor protein that is involved in cell migration, neurite outgrowth and vesicular transport. In multivesicular endosomes, Myosin Ib has been implicated in protein cargo traffic control.

REFERENCES

- 1. Marion, S., et al. 2005. Signalization and cytoskeleton activity through Myosin Ib during the early steps of phagocytosis in Entamoeba histolytica: a proteomic approach. Cell. Microbiol. 7: 1504-1518.
- 2. Salas-Cortes, L., et al. 2005. Myosin Ib modulates the morphology and the protein transport within multi-vesicular sorting endosomes. J. Cell Sci. 118: 4823-4832.
- 3. de Lanerolle, P., et al. 2005. Actin and Myosin I in the nucleus: what next? Nat. Struct. Mol. Biol. 12: 742-746.
- 4. Takeda, T., et al. 2005. Role of fission yeast Myosin I in organization of sterol-rich membrane domains. Curr. Biol. 15: 1331-1336.
- 5. Clark, R., et al. 2005. Loop 1 of transducer region in mammalian class I Myosin, Myosin Ib, modulates Actin affinity, ATPase activity, and nucleotide access. J. Biol. Chem. 280: 30935-30942.

CHROMOSOMAL LOCATION

Genetic locus: Myo1b (mouse) mapping to 1 C1.1.

PRODUCT

Myosin Ib shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10⁶ infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Myosin Ib siRNA (m): sc-44601 and Myosin Ib shRNA Plasmid (m): sc-44601-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

APPLICATIONS

Myosin Ib shRNA (m) Lentiviral Particles is recommended for the inhibition of Myosin Ib expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10⁶ infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Myosin Ib (F-8): sc-393053 is recommended as a control antibody for monitoring of Myosin Ib 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker[™] compatible goat antimouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Myosin Ib gene expression knockdown using RT-PCR Primer: Myosin Ib (m)-PR: sc-44601-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.