SANTA CRUZ BIOTECHNOLOGY, INC.

dystrophin siRNA (m): sc-35241

BACKGROUND

Dystrophin-glycoprotein complex (DGC) connects the F-Actin cytoskeleton on the inner surface of muscle fibers to the surrounding extracellular matrix, through the cell membrane interface. A deficiency in this protein contributes to Duchenne (DMD) and Becker (BMD) muscular dystrophies. The human dystrophin gene measures 2.4 megabases, has more than 80 exons, produces a 14 kb mRNA and contains at least eight independent tissue-specific promoters and two poly A sites. The dystrophin mRNA can undergo differential splicing and produce a range of transcripts that encode a large set of proteins. Dystrophin represents approximately 0.002% of total striated muscle protein and localizes to triadic junctions in skeletal muscle, where it is thought to influence calcium ion homeostasis and force transmission.

REFERENCES

1. Durbeej, M., et al. 2002. Muscular dystrophies involving the dystrophin-glycoprotein complex: an overview of current mouse models. Curr. Opin. Genet. Dev. 12: 349-361.
2. Michele, D.E., et al. 2003. Dystrophin-glycoprotein complex: post-translational processing and dystroglycan function. J. Biol. Chem. 278: 15457-15460.
3. Oak, S.A., et al. 2003. Skeletal muscle signaling pathway through the dystrophin glycoprotein complex and Rac 1. J. Biol. Chem. 278: 39287-39295.
4. Johnson, B.D., et al. 2005. Convergent regulation of skeletal muscle Ca^{2+} channels by dystrophin, the Actin cytoskeleton and cAMP-dependent protein kinase. Proc. Natl. Acad. Sci. USA 102: 4191-4196.
5. Bhasin, N., et al. 2005. Molecular extensibility of mini-dystrophins and a dystrophin rod construct. J. Mol. Biol. 352: 795-806.
6. Acharyya, S., et al. 2005. Dystrophin glycoprotein complex dysfunction: a regulatory link between muscular dystrophy and cancer cachexia. Cancer Cell 8: 421-432.
7. van der Plas, M.C., et al. 2006. Dystrophin is required for appropriate retrograde control of neurotransmitter release at the *Drosophila* neuromuscular junction. J. Neurosci. 26: 333-344.

CHROMOSOMAL LOCATION

Genetic locus: Dmd (mouse) mapping to X B.

PRODUCT

dystrophin 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 dystrophin shRNA Plasmid (m): sc-35241-SH and dystrophin shRNA (m) Lentiviral Particles: sc-35241-V as alternate gene silencing products.

For independent verification of dystrophin (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-35241A, sc-35241B and sc-35241C.

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

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

dystrophin (7A10): sc-47760 is recommended as a control antibody for monitoring of dystrophin 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 dystrophin gene expression knockdown using RT-PCR Primer: dystrophin (m)-PR: sc-35241-PR (20 µl, 509 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.