SANTA CRUZ BIOTECHNOLOGY, INC.

α/β-dystroglycan siRNA (m): sc-43489

BACKGROUND

Dystroglycan (DG) is a cell surface receptor for several extracellular matrix molecules including Laminins, Agrin and Perlecan. Dystroglycan function is required for the formation of basement membranes in early development and the organization of Laminin on the cell surface. α-dystroglycan is a membrane-associated, extracellular glycoprotein that is anchored to the cell-membrane by binding to the transmembrane glycoprotein β-dystroglycan to form an α/β-dystroglycan-complex. Additionally, dystroglycan is part of a multimolecular complex, where it associates with dystrophin, at the sarcolemma, to form the dystrophin-associated protein complex or with utrophin, at the neuromuscular junction, to form the utrophin-associated protein complex. Dystroglycan is also thought to participate in the clustering of nicotinic acetylcholine receptors at the neuromuscular junction.

REFERENCES

1. Cote, P.D., et al. 1999. Chimaeric mice deficient in dystroglycans develop muscular dystrophy and have disrupted myoneural synapses. Nat. Genet. 23: 338-342.
2. Seifert, J., et al. 2000. Syntheses of α-dystroglycan derived glycosyl amino acids carrying a novel mannosyl serine/threonine linkage. GIycoconj. J. 17: 407-423.
3. Henry, M.D., et al. 2001. Distinct roles for dystroglycan, β1 Integrin and perlecan in cell surface laminin organization. J. Cell Sci. 114: 1137-1144.
4. Masaki, T., et al. 2001. Expression of dystroglycan complex in satellite cells of dorsal root ganglia. Acta Neuropathol. 101: 174-178.
5. Marchand, S., et al. 2001. Differential targeting of components of the dystrophin complex to the postsynaptic membrane. Eur. J. Neurosci. 13: 221-229.
6. Bonuccelli, G., et al. 2007. Localized treatment with a novel FDA-approved proteasome inhibitor blocks the degradation of dystrophin and dystrophin-associated proteins in mdx mice. Cell Cycle. 6: 1242-1248.
7. Prados, B., et al. 2007. Expression of the murine Pomt1 gene in both the developing brain and adult muscle tissues and its relationship with clinical aspects of Walker-Warburg syndrome. Am. J. Pathol. 170: 1659-1668.

CHROMOSOMAL LOCATION

Genetic locus: Dag1 (mouse) mapping to 9 F2.

PRODUCT

α/β-dystroglycan 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 α/β-dystroglycan shRNA Plasmid (m): sc-43489-SH and α/β-dystroglycan shRNA (m) Lentiviral Particles: sc-43489-V as alternate gene silencing products.

For independent verification of α/β-dystroglycan (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-43489A, sc-43489B and sc-43489C.

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

α/β-dystroglycan siRNA (m) is recommended for the inhibition of α/β-dystroglycan 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

α-dystroglycan (IIH6): sc-53987 is recommended as a control antibody for monitoring of α/β-dystroglycan 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 α/β-dystroglycan gene expression knockdown using RT-PCR Primer: α/β-dystroglycan (m)-PR: sc-43489-PR (20 µl, 497 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.