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Laminin α -2 siRNA (h): sc-43143

BACKGROUND

Laminins are essential and abundant structural non-collagenous glycoproteins localizing to basement membranes. Basement membranes (cell-associated extracellular matrices (ECMs)) are polymers of laminins with stabilizing Type IV collagen networks, Nidogen and several proteoglycans. Basement membranes are found under epithelial layers, around the endothelium of blood vessels, and surrounding muscle, peripheral nerve and fat cells. Formation of basement membranes influences cell proliferation, phenotype, migration, gene expression and tissue architecture. Each Laminin is a heterotrimer of α , β , and γ chain subunits that undergoes cell-secretion and incorporation into the ECM. Laminins can self-assemble, bind to other matrix macromolecules, and have unique and shared cell interactions mediated by integrins, dystroglycan and cognate Laminin receptors. The human Laminin α -2 gene is necessary for sustenance of mature muscle cells. The Laminin α -2 gene is associated with congenital muscular dystrophy (CMD) in humans and dystrophia muscularis in mice.

REFERENCES

1. Tryggvason, K. 1993. The Laminin family. *Curr. Opin. Cell Biol.* 5: 877-882.
2. Schnaper, H.W., et al. 1993. Role of Laminin in endothelial cell recognition and differentiation. *Kidney Int.* 43: 20-25.
3. Engvall, E., et al. 1996. Domains of Laminin. *J. Cell. Biochem.* 61: 493-501.
4. Kuang, W., et al. 1998. Disruption of the LAMA2 gene in embryonic stem cells: Laminin α -2 is necessary for sustenance of mature muscle cells. *Exp. Cell Res.* 241: 117-125.
5. Ekblom, M., et al. 1998. Laminin isoforms and epithelial development. *Ann. N.Y. Acad. Sci.* 857: 194-211.

CHROMOSOMAL LOCATION

Genetic locus: LAMA2 (human) mapping to 6q22.33.

PRODUCT

Laminin α -2 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Laminin α -2 shRNA Plasmid (h): sc-43143-SH and Laminin α -2 shRNA (h) Lentiviral Particles: sc-43143-V as alternate gene silencing products.

For independent verification of Laminin α -2 (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-43143A, sc-43143B and sc-43143C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Laminin α -2 siRNA (h) is recommended for the inhibition of Laminin α -2 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 μ M in 66 μ l. 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

Laminin α -2 (B-4): sc-55605 is recommended as a control antibody for monitoring of Laminin α -2 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 Laminin α -2 gene expression knockdown using RT-PCR Primer: Laminin α -2 (h)-PR: sc-43143-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.