SANTA CRUZ BIOTECHNOLOGY, INC.

smoothelin siRNA (m): sc-36513

BACKGROUND

The cytoskeletal protein smoothelin is highly conserved among vertebrates and is expressed exclusively by contractile smooth muscle cells where it localizes to the filament network. Smoothelin associates with actin stress fibers but does not interact with desmin. At least two isoforms of smoothelin are produced by alternative splicing. The short isoform lacks amino acids 1-544 at the amino-terminus of the long isoform. The short isoform is expressed in visceral muscle tissue, including intestine and stomach, but not in brain, while the long isoform is expressed in all vascularized organs. In the vascular system, smoothelin expression is limited to large veins and arteries capable of pulsatile contraction. As a marker for the highly differentiated contractile smooth muscle cell, smoothelin expression is useful for studying vascular malformation and injury. The gene encoding human smoothelin maps to chromosome 22q12.2.

REFERENCES

1. van der Loop, F., et al. 1996. Smoothelin, a novel cytoskeletal protein specific for smooth muscle cells. J. Cell Biol. 34: 401-411.
2. van Eys, G., et al. 1997. Smoothelin expression characteristics: development of a smooth muscle cell *in vitro* system and identification of a vascular variant. Cell Struct. Funct. 22: 65-72.
3. van der Loop, F., et al. 1997. Differentiation of smooth muscle cells in human blood vessels as defined by smoothelin, a novel marker for the contractile phenotype. Arterioscler. Thromb. Vasc. Biol. 17: 665-671.
4. Johnson, J., et al. 2001. Injury induces dedifferentiation of smooth muscle cells and increased matrix-degrading metalloproteinase activity in human saphenous vein. Arterioscler. Thromb. Vasc. Biol. 21: 1146-1151.
5. Rensen, S., et al. 2002. Expression of the smoothelin gene is mediated by alternative promoters. Cardiovasc. Res. 55: 850-863.
6. Maeng, M., et al. 2003. Adventitial myofibroblasts play no major role in neointima formation after angioplasty. Scand. Cardiovasc. J. 37: 34-42.

CHROMOSOMAL LOCATION

Genetic locus: Smtn (mouse) mapping to 11 A1.

PRODUCT

smoothelin 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 smoothelin shRNA Plasmid (m): sc-36513-SH and smoothelin shRNA (m) Lentiviral Particles: sc-36513-V as alternate gene silencing products.

For independent verification of smoothelin (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-36513A, sc-36513B and sc-36513C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

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

smoothelin (C-8): sc-376902 is recommended as a control antibody for monitoring of smoothelin 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 smoothelin gene expression knockdown using RT-PCR Primer: smoothelin (m)-PR: sc-36513-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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