SANTA CRUZ BIOTECHNOLOGY, INC.

I-Plastin siRNA (h): sc-78197

BACKGROUND

Smooth muscle is present in many areas throughout the body, including vascular, respiratory, urinary and gastrointestinal tissue, and its ability to contract is largely due to calcium gradients. I-Plastin (intestine-specific Plastin) is also known as PLS1 (Plastin-1) or Fimbrin and is a 629 amino acid protein that is abundantly expressed in small intestine, colon and kidneys and is also expressed at lower concentrations in lungs and stomach. II-Plastin localizes to the brush border and more specifically to the cytoplasm of cells. I-Plastin is a member of the plastin family of Actin-binding proteins. The ability of I-Plastin to bind Actin allows it to collect Actin filaments into bundles in the absence of calcium, which, when present, is thought to regulate or inhibit Actin bundling. I-Plastin contains four CH (calponin homology) domains that are thought to bind Actin, unless they are phosphorylated. The N-terminus of I-Plastin has a calmodulin-like calcium-binding domain followed by two α-actinin-like Actin binding domains that also bind calcium. I-Plastin also contains two EF hand domains important for calcium binding and protein-protein interactions. I-Plastin is thought to cross-link Actin filaments and to have an inhibitory relationship with calcium, but the extent to which this occurs is not known.

REFERENCES

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2. Lin, C.S., et al. 1993. Human plastin genes. Comparative gene structure, chromosome location, and differential expression in normal and neoplastic cells. J. Biol. Chem. 268: 2781-2792.
3. Lin, C.S., et al. 1994. Identification of I-plastin, a human fimbrin isoform expressed in intestine and kidney. Mol. Cell. Biol. 14: 2457-2467.
4. Horowitz, A., et al. 1996. Mechanisms of smooth muscle contraction. Physiol. Rev. 76: 967-1003.
5. Online Mendelian Inheritance in Man, OMIM™. 1998. Johns Hopkins University, Baltimore, MD. MIM Number: 602734. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
6. Lewit-Bentley, A., et al. 2000. EF-hand calcium-binding proteins. Curr. Opin. Struct. Biol. 10: 637-643.

CHROMOSOMAL LOCATION

Genetic locus: PLS1 (human) mapping to 3q23.

PRODUCT

I-Plastin 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see I-Plastin shRNA Plasmid (h): sc-78197-SH and I-Plastin shRNA (h) Lentiviral Particles: sc-78197-V as alternate gene silencing products.

For independent verification of I-Plastin (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-78197A, sc-78197B and sc-78197C.

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

I-Plastin siRNA (h) is recommended for the inhibition of I-Plastin 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 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

Plastin (F-2): sc-271223 is recommended as a control antibody for monitoring of I-Plastin 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 I-Plastin gene expression knockdown using RT-PCR Primer: I-Plastin (h)-PR: sc-78197-PR (20 µl). 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.