SANTA CRUZ BIOTECHNOLOGY, INC.

Caldesmon siRNA (m): sc-29881

BACKGROUND

Caldesmon, Filamin 1, Nebulin and Villin are differentially expressed and regulated Actin binding proteins. Both muscular and non-muscular forms of Caldesmon have been identified and each has been shown to bind to Actin as well as to calmodulin and Myosin. Alternative splicing of the gene encoding Caldesmon results in five isoforms. Muscular Caldesmon (isoform 1), also designated high molecular weight Caldesmon or H-Caldesmon (H-CAD), is expressed predominantly on thin filaments in smooth muscle. Non-muscular Caldesmon (isoforms 2-5), also designated low molecular weight Caldesmon or L-Caldesmon (L-CAD), is widely expressed in non-muscle tissues and cells. Filamin 1, which is ubiquitously expressed and exists as a homodimer, functions to crosslink Actin to filaments. Nebulin is a large filamentous protein specific to muscle tissue that may function as a ruler for filament length. Several isoforms of Nebulin are produced by alternative exon usage. Villin is Ca^{2+}-regulated and is the major structural component of the brush border of absorptive cells.

REFERENCES

1. Weihing, R.R. 1988. Actin-binding and dimerization domains of HeLa cell Filamin. Biochemistry 27: 1865-1869.
2. Marston, S., et al. 1992. Caldesmon binds to smooth muscle myosin and myosin rod and crosslinks thick filaments to actin filaments. J. Muscle Res. Cell Motil. 13: 206-218.
3. Maunoury, R., et al. 1992. Developmental regulation of Villin gene expression in the epithelial cell lineages of mouse digestive and urogenital tracts. Development 115: 717-728.
4. Labeit, S., et al. 1995. The complete primary structure of human nebulin and its correlation to muscle structure. J. Mol. Biol. 248: 308-315.
5. Zhang, J.Q., et al. 1996. cDNA cloning of mouse nebulin. Evidence that the Nebulin-coding sequence is highly conserved among vertebrates. Eur. J. Biochem. 239: 835-841.
6. Huber, P.A., et al. 1996. Multiple-sited interaction of Caldesmon with Ca^{2+}-calmodulin. Biochem. J. 316: 413-420.

CHROMOSOMAL LOCATION

Genetic locus: Cald1 (mouse) mapping to 6 B1.

PRODUCT

Caldesmon 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 Caldesmon shRNA Plasmid (m): sc-29881-SH and Caldesmon shRNA (m) Lentiviral Particles: sc-29881-V as alternate gene silencing products.

For independent verification of Caldesmon (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-29881A, sc-29881B and sc-29881C.

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

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

Caldesmon (C21): sc-58700 is recommended as a control antibody for monitoring of Caldesmon 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 Caldesmon gene expression knockdown using RT-PCR Primer: Caldesmon (m)-PR: sc-29881-PR (20 µl, 417 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Liou, Y.M., et al. 2018. Effect of l-caldesmon on osteoclastogenesis in RANKL-induced RAW264.7 cells. J. Cell. Physiol. 233: 6888-6901.

RESEARCH USE

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