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CapG shRNA (m) Lentiviral Particles: sc-44921-V

BACKGROUND

Caldesmon, Filamin 1, Nebulin, Plastin, ADF, Gelsolin, CapG, Dematin and Cofilin are differentially expressed Actin-binding proteins. Both muscular (CDh) and non-muscular (CD1) forms of Caldesmon bind to Actin as well as to Calmodulin and Myosin. CDh is expressed predominantly on thin filaments in smooth muscle, whereas CD1 is widely expressed in non-muscle tissues and cells. CapG, also designated Actin-regulatory protein and macrophage-capping protein, is a macrophage-specific protein that reversibly blocks the barbed ends of Actin filaments, but does not sever preformed ones. The interactions of CapG with Actin may be important in the regulation of nuclear and cytoplasmic structures. CapG is a calcium-sensitive DNA-binding protein that plays a role in macrophage function. It is expressed in macrophages and macrophage-like cells and can localize both to the nucleus and the cytoplasm.

REFERENCES

1. Dabiri, G.A., et al. 1992. Molecular cloning of human macrophage-capping protein cDNA. A unique member of the Gelsolin/Villin family expressed primarily in macrophages. *J. Biol. Chem.* 267: 16545-16552.
2. Mishra, V.S., et al. 1994. The human Actin-regulatory protein CapG: gene structure and chromosome location. *Genomics* 23: 560-565.
3. Southwick, F.S., et al. 1995. Gain-of-function mutations conferring Actin-severing activity to human macrophage CapG. *J. Biol. Chem.* 270: 45-48.
4. Pellieux, C., et al. 2003. CapG, a Gelsolin family protein modulating protective effects of unidirectional shear stress. *J. Biol. Chem.* 278: 29136-29144.
5. SWISS-PROT/TrEMBL (P40121). World Wide Web URL: <http://www.expasy.ch/sprot/sprot-top.html>

CHROMOSOMAL LOCATION

Genetic locus: Capg (mouse) mapping to 6 C1.

PRODUCT

CapG shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see CapG siRNA (m): sc-44921 and CapG shRNA Plasmid (m): sc-44921-SH as alternate gene silencing products.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

APPLICATIONS

CapG shRNA (m) Lentiviral Particles is recommended for the inhibition of CapG expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

CapG (C-20): sc-33084 is recommended as a control antibody for monitoring of CapG 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CapG gene expression knockdown using RT-PCR Primer: CapG (m)-PR: sc-44921-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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