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Myocardin shRNA (h) Lentiviral Particles: sc-43953-V



The Power to Question

BACKGROUND

Serum response factor (SRF) is a transcription factor that binds the serum response element (SRE), a sequence that mediates the transient response of many cellular genes to growth stimulation. SRF-binding sites are also constitutive promoter elements in many muscle-specific promoters. Myocardin associates with SRF in cardiac muscle cells to activate cardiac muscle promoters. Myocardin is also expressed in smooth muscle cells and appears to play a role in cell differentiation. Specifically, myocardin is expressed in vascular smooth muscle within the aortic arteries and pulmonary outflow tract as well as in the genitourinary tract and gastrointestinal tract. Myocardin is absent in the coronary vasculature, dorsal aorta, skeletal muscle and other non-muscle tissue types. Myocardin belongs to the SAP (SAF-A/B, Acinus and PIAS) domain family of nuclear proteins which includes hnRNP U and PIAS. The SAP domain may play a role in targeting proteins to specific chromosomal locations.

REFERENCES

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- Boxer, L.M., et al. 1989. The sarcomeric actin CArG-binding factor is indistinguishable from the c-fos serum response factor. Mol. Cell. Biol. 9: 515-522.
- 3. Treisman, R. 1990. The SRE: a growth factor responsive transcriptional regulator. Sem. Cancer Biol. 1: 47-58.
- Hill, C.S., et al. 1993. Functional analysis of a growth factor-responsive transcription factor complex. Cell 73: 395-406.
- Aravind, L., et al. 2000. SAP-a putative DNA-binding motif involved in chromosomal organization. Trends Biochem. Sci. 25: 112-114.
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CHROMOSOMAL LOCATION

Genetic locus: MYOCD (human) mapping to 17p12.

PRODUCT

Myocardin shRNA (h) 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 μl frozen stock containing 1.0 x 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 Myocardin siRNA (h): sc-43953 and Myocardin shRNA Plasmid (h): sc-43953-SH as alternate gene silencing products.

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.

APPLICATIONS

Myocardin shRNA (h) Lentiviral Particles is recommended for the inhibition of Myocardin expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0 x 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

Myocardin (A-13): sc-34238 is recommended as a control antibody for monitoring of Myocardin 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 Myocardin gene expression knockdown using RT-PCR Primer: Myocardin (h)-PR: sc-43953-PR (20 μ I, 425 bp). 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.

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.

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