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HMGR shRNA (h) Lentiviral Particles: sc-43838-V

BACKGROUND

The human enzyme hydroxy-3-methylglutaryl coenzyme A reductase (HMGR) limits the rate of cholesterol synthesis, a necessary process for cellular growth, in liver tissue. Phosphorylation of HMGR inactivates the enzyme, which occurs via a negative feedback mechanism mediated by sterols and non-sterol metabolites derived from the product of the reductase reaction. Inhibitors of HMGR (statins) exert anti-inflammatory effects and decrease the frequency of cardiovascular events by lowering plasma cholesterol. Additionally, intermediate products along the pathway catalyzed by HMGR, which modulate signal transducing proteins such as Ras, provide possible ties between HMGR regulation and new chemotherapeutic methods.

REFERENCES

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2. Duhamel-Clerin, E., et al. 1994. Cellular expression of an HMGR promoter-CAT fusion gene in transgenic mouse brain: evidence for a developmental regulation in oligodendrocytes. *Glia* 11: 35-46
3. Zager, R.A., et al. 2002. The mevalonate pathway during acute tubular injury: selected determinants and consequences. *Am. J. Pathol.* 161: 681-692.
4. Viedt, C., et al. 2003. HMG-CoA reductase inhibition reduces the proinflammatory activation of human vascular smooth muscle cells by the terminal complement factor C5b-9. *Basic Res. Cardiol.* 98: 353-361.
5. Wassmann, S., et al. 2003. Rapid effect of 3-hydroxy-3-methylglutaryl coenzyme a reductase inhibition on coronary endothelial function. *Circ. Res.* 93: 98-103.
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CHROMOSOMAL LOCATION

Genetic locus: HMGR (human) mapping to 5q13.3.

PRODUCT

HMGR 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 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 HMGR siRNA (h): sc-43838 and HMGR shRNA Plasmid (h): sc-43838-SH as alternate gene silencing products.

APPLICATIONS

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

PROTOCOLS

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

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

HMGR (C-1): sc-271595 is recommended as a control antibody for monitoring of HMGR 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 goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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 goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor HMGR gene expression knockdown using RT-PCR Primer: HMGR (h)-PR: sc-43838-PR (20 μ l, 538 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.

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.

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.