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HMGCR siRNA (m): sc-44851

BACKGROUND

The human enzyme hydroxy-3-methylglutaryl coenzyme A reductase (HMGCR) limits the rate of cholesterol synthesis, a necessary process for cellular growth, in liver tissue. Phosphorylation of HMGCR 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 HMGCR (statins) exert anti-inflammatory effects and decrease the frequency of cardiovascular events by lowering plasma cholesterol. Additionally, intermediate products along the pathway catalyzed by HMGCR, which modulate signal transducing proteins such as Ras, provide possible ties between HMGCR regulation and new chemotherapeutic methods.

REFERENCES

1. Luskey, K.L., et al. 1985. Human 3-hydroxy-methylglutaryl coenzyme A reductase. *J. Biol. Chem.* 260: 10271-10277.
2. Duhamel-Clerin, E., et al. 1994. Cellular expression of an HMGCR 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.

CHROMOSOMAL LOCATION

Genetic locus: *Hmgcr* (mouse) mapping to 13 D1.

PRODUCT

HMGCR 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HMGCR shRNA Plasmid (m): sc-44851-SH and HMGCR shRNA (m) Lentiviral Particles: sc-44851-V as alternate gene silencing products.

For independent verification of HMGCR (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-44851A, sc-44851B and sc-44851C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

HMGCR siRNA (m) is recommended for the inhibition of HMGCR 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 μ M in 66 μ l. 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

HMGCR (C-1): sc-271595 is recommended as a control antibody for monitoring of HMGCR 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 HMGCR gene expression knockdown using RT-PCR Primer: HMGCR (m)-PR: sc-44851-PR (20 μ l, 573 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Rub, A., et al. 2009. Cholesterol depletion associated with *Leishmania major* infection alters macrophage CD40 signalosome composition and effector function. *Nat. Immunol.* 10: 273-280.

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.