

apoB siRNA (h): sc-41180

BACKGROUND

Post-transcriptional editing of apolipoprotein B (apoB) mRNA is regulated by APOBEC1 (also designated human (or rat) small intestinal apolipoprotein B mRNA editing protein, HEPR, or REPR) in hepatic cells to achieve a steady state proportion of edited and unedited RNA molecules. Two forms of apoB are known to circulate in the plasma of mammals. apoB-100 is a protein primarily synthesized in the liver as a structural component of very-low-density lipoprotein particles. A truncated form of apoB-100, apoB-48, is synthesized in the small intestine and contains the amino-terminal 2,152 amino acids of the larger protein. This organ-specific partitioning of apoB production is the result of RNA editing of a common apoB gene.

REFERENCES

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2. Law, S.W., et al. 1986. Human liver apolipoprotein B-100 cDNA: complete nucleic acid and derived amino acid sequence. *Proc. Natl. Acad. Sci. USA* 83: 8142-8146.
3. Young, S.G., et al. 1986. Two new monoclonal antibody-based enzyme-linked assays of apolipoprotein B. *Clin. Chem.* 32: 1484-1490.
4. Micic, S., et al. 1989. A-I and B in blood spotted on filter paper. *Clin. Chem.* 34: 2452-2455.
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6. Anant, S., et al. 2000. An AU-rich sequence element (UUUN[A/U]U) downstream of the edited C in apolipoprotein B mRNA is a high-affinity binding site for APOBEC1: binding of APOBEC1 to this motif in the 3' untranslated region of c-Myc increases mRNA stability. *Mol. Cell. Biol.* 20: 1982-1992.
7. Yang, Y., et al. 2000. Induction of cytidine to uridine editing on cytoplasmic apolipoprotein B mRNA by overexpressing Apobec-1. *J. Biol. Chem.* 275: 22663-22669.
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CHROMOSOMAL LOCATION

Genetic locus: APOB (human) mapping to 2p24.1.

PRODUCT

apoB siRNA (h) 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 apoB shRNA Plasmid (h): sc-41180-SH and apoB shRNA (h) Lentiviral Particles: sc-41180-V as alternate gene silencing products.

For independent verification of apoB (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-41180A, sc-41180B and sc-41180C.

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

apoB siRNA (h) is recommended for the inhibition of apoB expression in human 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

apoB (C1.4): sc-13538 is recommended as a control antibody for monitoring of apoB 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 apoB gene expression knockdown using RT-PCR Primer: apoB (h)-PR: sc-41180-PR (20 μ l, 455 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

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.