

apoH siRNA (m): sc-72519

BACKGROUND

Human apolipoprotein H (apoH, also designated β_2 -glycoprotein I, activated protein C binding protein or APC inhibitor) is a five-domain plasma membrane-adhesion protein that is rich in sialic acid linked to galactose or N-acetylglactosamine. ApoH has been implicated in a variety of physiological pathways, including blood coagulation and the immune response. ApoH is a cofactor for the binding of serum auto-antibodies from antiphospholipid syndrome, and is correlated with thrombosis, lupus erythematosus and recurrent fetal loss. In addition, apoH is also implicated in the clearance of apoptotic bodies from the circulation. The apoH gene is located on human chromosome 17q24.2. ApoH is synthesized by hepatocytes and is present in blood associated with plasma lipoproteins. ApoH displays a genetically determined structural polymorphism including three alleles (apoH*1, apoH*2 and apoH*3). ApoH can inhibit the translocation of cholesterol from extracellular pools to macrophages, which reduces the cellular accumulation of cholesterol, suggesting that apoH may play an important role in the prevention of atherosclerosis.

REFERENCES

1. Mehdi, H., et al. 1991. Nucleotide sequence and expression of the human gene encoding apolipoprotein H (β_2 -glycoprotein I). *Gene* 108: 293-298.
2. Steinkasserer, A., et al. 1991. Complete nucleotide and deduced amino acid sequence of human β_2 -glycoprotein I. *Biochem. J.* 277: 387-391.
3. Ruii, G., et al. 1997. Influence of apoH protein polymorphism on apoH levels in normal and diabetic subjects. *Clin. Genet.* 52: 167-172.
4. Gambino, R., et al. 1997. Qualitative analysis of the carbohydrate composition of apolipoprotein H. *J. Protein Chem.* 16: 205-212.
5. Bouma, B., et al. 1999. Adhesion mechanism of human β_2 -glycoprotein I to phospholipids based on its crystal structure. *EMBO J.* 18: 5166-5174.
6. Schwarzenbacher, R., et al. 1999. Crystal structure of human β_2 -glycoprotein I: implications for phospholipid binding and the antiphospholipid syndrome. *EMBO J.* 18: 6228-6239.

CHROMOSOMAL LOCATION

Genetic locus: Apoh (mouse) mapping to 11 E1.

PRODUCT

apoH 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 apoH shRNA Plasmid (m): sc-72519-SH and apoH shRNA (m) Lentiviral Particles: sc-72519-V as alternate gene silencing products.

For independent verification of apoH (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-72519A, sc-72519B and sc-72519C.

PROTOCOLS

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

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

apoH siRNA (m) is recommended for the inhibition of apoH 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

apoH (C-2): sc-515677 is recommended as a control antibody for monitoring of apoH 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 apoH gene expression knockdown using RT-PCR Primer: apoH (m)-PR: sc-72519-PR (20 μ l). 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.