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MTP siRNA (h): sc-45275

BACKGROUND

Microsomal triglyceride transfer protein (MTP) catalyzes the transport of cholesteryl ester, triglyceride and phospholipid between phospholipid surfaces. MTP is a heterodimer consisting of MTP and PDI (protein disulfide isomerase). It is required for the secretion of plasma lipoproteins containing apolipoprotein B. It is negatively regulated by Insulin and positively regulated by cholesterol. MTP, which localizes to the endoplasmic reticulum (ER), is expressed primarily in small intestine, liver, kidney, testis and ovary. It is not expressed in epithelial cells. Defects in the MTP gene can cause abetalipoproteinemia (ABL) which is an autosomal recessive lipoprotein metabolism disorder.

REFERENCES

1. Shoulders, C.C., et al. 1994. The abetalipoproteinemia gene is a member of the vitellogenin family and encodes an α -helical domain. *Nat. Struct. Biol.* 1: 285-286.
2. Hagan, D.L., et al. 1994. Transcriptional regulation of human and hamster microsomal triglyceride transfer protein genes. Cell type-specific expression and response to metabolic regulators. *J. Biol. Chem.* 269: 28737-28744.
3. Rehberg, E.F., et al. 1996. A novel abetalipoproteinemia genotype. Identification of a missense mutation in the 97 kDa subunit of the microsomal triglyceride transfer protein that prevents complex formation with protein disulfide isomerase. *J. Biol. Chem.* 271: 29945-29952.
4. Ohashi, K., et al. 2000. Novel mutations in the microsomal triglyceride transfer protein gene causing abetalipoproteinemia. *J. Lipid Res.* 41: 1199-1204.
5. Wang, J., et al. 2000. Microsomal triglyceride transfer protein (MTP) gene mutations in Canadian subjects with abetalipoproteinemia. *Hum. Mutat.* 15: 294-295.

CHROMOSOMAL LOCATION

Genetic locus: MTTP (human) mapping to 4q23.

PRODUCT

MTP 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 MTP shRNA Plasmid (h): sc-45275-SH and MTP shRNA (h) Lentiviral Particles: sc-45275-V as alternate gene silencing products.

For independent verification of MTP (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-45275A, sc-45275B and sc-45275C.

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

MTP siRNA (h) is recommended for the inhibition of MTP 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

MTP (C-1): sc-515742 is recommended as a control antibody for monitoring of MTP 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 MTP gene expression knockdown using RT-PCR Primer: MTP (h)-PR: sc-45275-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.