

CPTII siRNA (h): sc-40378

BACKGROUND

The mitochondrial β -oxidation of long-chain fatty acids is initiated by the sequential action of carnitine palmitoyltransferase (CPT) I (outer membrane and detergent labile) and II (inner membrane and detergent stable), together with carnitine carrier. CPTI catalyzes the first reaction in the transport of long-chain fatty acids from the cytoplasm to the mitochondrion, a rate-limiting step in β -oxidation. Two types of CPTI are known, the liver (CPTIA) and muscle (CPTIB) isoforms. The muscle type protein is specially expressed in heart and skeletal muscle. Membrane-bound CPTI, but not CPTII, is inhibited reversibly by malonyl-coenzyme A (CoA). Unlike CPTII, CPTI requires membrane integrity for catalytic function. In addition, glutamic acid 3 and histidine 5 are necessary for malonyl CoA inhibition and binding to liver CPTI, but not for catalytic activity.

REFERENCES

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4. Britton, C.H., et al. 1995. Human liver mitochondrial carnitine palmitoyltransferase I: characterization of its cDNA and chromosomal localization and partial analysis of the gene. *Proc. Natl. Acad. Sci. USA* 92: 1984-1988.
5. Yamazaki, N., et al. 1996. Isolation and characterization of cDNA and genomic clones encoding human muscle type carnitine palmitoyltransferase I. *Biochim. Biophys. Acta* 1307: 157-161.
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CHROMOSOMAL LOCATION

Genetic locus: CPT2 (human) mapping to 1p32.3.

PRODUCT

CPTII 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 CPTII shRNA Plasmid (h): sc-40378-SH and CPTII shRNA (h) Lentiviral Particles: sc-40378-V as alternate gene silencing products.

For independent verification of CPTII (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-40378A, sc-40378B and sc-40378C.

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

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

CPTII (G-5): sc-377294 is recommended as a control antibody for monitoring of CPTII 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 CPTII gene expression knockdown using RT-PCR Primer: CPTII (h)-PR: sc-40378-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.

PROTOCOLS

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