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OCTN1 siRNA (h): sc-42558

BACKGROUND

Carnitine (β -hydroxy- γ -trimethylaminobutyrate) is a small, highly polar compound that aids in the β -oxidation of long-chain fatty acids. Organic cation/carnitine transporters (OCTN) assist in the elimination of cationic compounds, including xenobiotics, and transport carnitine for reabsorption in the kidney. Similar to organic cation transporters (OCT), OCTN proteins localize to the plasma membrane of epithelial cells. OCTN1 is expressed in kidney, trachea, bone marrow and fetal liver. OCTN2 is abundantly expressed in kidney, skeletal muscle, placenta and heart. OCTN3 is strongly expressed in testis and weakly expressed in kidney. The gene encoding human OCTN1 maps to chromosome 5 and the gene encoding human OCTN2 maps to chromosome 5q31. Mutations in the gene encoding OCTN2 leads to systemic carnitine deficiency (SCD), an autosomal recessive disorder characterized by cardiomyopathy, skeletal myopathy, lethargy, hypoglycemia and hyperammonemia.

REFERENCES

1. Tamai, I., et al. 1998. Molecular and functional identification of sodium ion-dependent, high affinity human carnitine transporter OCTN2. *J. Biol. Chem.* 273: 20378-20382.
2. Wu, X., et al. 1998. cDNA sequence, transport function, and genomic organization of human OCTN2, a new member of the organic cation transporter family. *Biochem. Biophys. Res. Commun.* 246: 589-595.
3. Lamhonwah, A.M., et al. 1998. Carnitine uptake defect: frameshift mutations in the human plasmalemmal carnitine transporter gene. *Biochem. Biophys. Res. Commun.* 252: 396-401.
4. Km, L., et al. 1998. A missense mutation of mouse OCTN2, a sodium-dependent carnitine cotransporter, in the juvenile visceral steatosis mouse. *Biochem. Biophys. Res. Commun.* 252: 590-594.

CHROMOSOMAL LOCATION

Genetic locus: SLC22A4 (human) mapping to 5q31.1.

PRODUCT

OCTN1 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 OCTN1 shRNA Plasmid (h): sc-42558-SH and OCTN1 shRNA (h) Lentiviral Particles: sc-42558-V as alternate gene silencing products.

For independent verification of OCTN1 (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-42558A, sc-42558B and sc-42558C.

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.

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

OCTN1 siRNA (h) is recommended for the inhibition of OCTN1 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

OCTN1/2 (H-9): sc-515731 is recommended as a control antibody for monitoring of OCTN1 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 OCTN1 gene expression knockdown using RT-PCR Primer: OCTN1 (h)-PR: sc-42558-PR (20 μ l, 427 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.