

MCAT siRNA (h): sc-75760

BACKGROUND

The biosynthesis of fatty acids can occur in the cytoplasm, via the type I fatty acid synthase complex, or in mitochondria, via the type II malonyl-CoA-dependent system. MCAT (mitochondrial malonyl CoA:ACP acyltransferase), also known as MT, MCT (mitochondrial malonyltransferase), fabD or FASN2C, is a member of the type II malonyltransferase family of proteins. Localizing to mitochondria, MCAT is encoded by a nuclear gene and, via an N-terminal localization signal, it is subsequently imported into mitochondria. MCAT functions in lipid metabolism and may be a component of a mitochondrial fatty acid synthase complex. More specifically, MCAT catalyzes the transfer of a malonyl group from malonyl-CoA to the mitochondrial acyl carrier protein (NDUFAB1), a subunit of respiratory complex 1. This reaction is essential in the initiation of the type II fatty acid biosynthesis system. Two isoforms of MCAT exist due to alternative splicing events.

REFERENCES

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- Kastaniotis, A.J., Autio, K.J., Sormunen, R.T. and Hiltunen, J.K. 2004. Htd2p/Yhr067p is a yeast 3-hydroxyacyl-ACP dehydratase essential for mitochondrial function and morphology. *Mol. Microbiol.* 53: 1407-1421.
- Maier, T., Jenni, S. and Ban, N. 2006. Architecture of mammalian fatty acid synthase at 4.5 Å resolution. *Science* 311: 1258-1262.
- Zhang, L., Liu, W., Xiao, J., Hu, T., Chen, J., Chen, K., Jiang, H. and Shen, X. 2007. Malonyl-CoA: acyl carrier protein transacylase from *Helicobacter pylori*: crystal structure and its interaction with acyl carrier protein. *Protein Sci.* 16: 1184-1192.

CHROMOSOMAL LOCATION

Genetic locus: MCAT (human) mapping to 22q13.2.

PRODUCT

MCAT 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 MCAT shRNA Plasmid (h): sc-75760-SH and MCAT shRNA (h) Lentiviral Particles: sc-75760-V as alternate gene silencing products.

For independent verification of MCAT (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-75760A, sc-75760B and sc-75760C.

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

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

MCAT (E-11): sc-390858 is recommended as a control antibody for monitoring of MCAT 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 MCAT gene expression knockdown using RT-PCR Primer: MCAT (h)-PR: sc-75760-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.