

ETFDH siRNA (h): sc-89048

BACKGROUND

ETFDH (electron-transferring-flavoprotein dehydrogenase), also known as electron transfer flavoprotein-ubiquinone oxidoreductase, MADD or ETFQO, is a 617 amino acid membrane-bound electron transfer protein that exists as a monomer, localizes to the mitochondrial inner membrane and belongs to the ETF-QO/fixC family. ETFDH accepts electrons from electron-transfer flavoprotein (ETF) in the mitochondrial matrix while reducing ubiquinone in the mitochondrial membrane. ETFDH is encoded by a gene mapping to human chromosome 4q32.1, and contains one molecule of FAD and a 4Fe-4S cluster. As a result of alternative splicing events, two ETFDH isoforms exist. Defects in ETFDH are responsible for an autosomal recessive disorder of amino acid, fatty acid and choline metabolism, known as glutaric aciduria type 2C (GA2C) or multiple acyl-CoA dehydrogenation deficiency (MADD). GA2C is characterized by severe hypoketotic hypoglycemia and acidosis.

REFERENCES

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CHROMOSOMAL LOCATION

Genetic locus: ETFDH (human) mapping to 4q32.1.

PRODUCT

ETFDH 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 ETFDH shRNA Plasmid (h): sc-89048-SH and ETFDH shRNA (h) Lentiviral Particles: sc-89048-V as alternate gene silencing products.

For independent verification of ETFDH (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-89048A, sc-89048B and sc-89048C.

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

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

ETFDH (D-2): sc-515202 is recommended as a control antibody for monitoring of ETFDH 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 ETFDH gene expression knockdown using RT-PCR Primer: ETFDH (h)-PR: sc-89048-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.