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ETFB siRNA (m): sc-144954

BACKGROUND

ETFB (electron transfer flavoprotein subunit beta), also known as FP585, MADD or β -ETF, is a 255 amino acid protein that belongs to the ETF β -subunit/fixA family. Localizing to the mitochondrion matrix, ETFB is abundantly expressed in liver, heart and skeletal muscle, with weaker levels of expression found in brain, placenta, lung, kidney and pancreas. ETFB exists as a heterodimer of an alpha and beta subunit; this dimer utilizes FAD as a cofactor and binds one AMP per subunit. The gene encoding ETFB maps to human chromosome 19q13.33 and mouse chromosome 7 B4. Defects to this gene have been linked to glutaric aciduria type 2B (GA2B), an autosomal recessive disorder of fatty acid, amino acid and choline metabolism. ETFB acts as a shuttle for electrons, transferring them between primary flavoprotein dehydrogenases and the membrane-bound electron transfer flavoprotein ubiquinone oxidoreductase.

REFERENCES

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- Finocchiaro, G., et al. 1993. cDNA cloning and mitochondrial import of the β -subunit of the human electron-transfer flavoprotein. *Eur. J. Biochem.* 213: 1003-1008.
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- Colombo, I., et al. 1994. Mutations and polymorphisms of the gene encoding the β -subunit of the electron transfer flavoprotein in three patients with glutaric acidemia type II. *Hum. Mol. Genet.* 3: 429-435.
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- Roberts, D.L., et al. 1996. Three-dimensional structure of human electron transfer flavoprotein to 2.1-Å resolution. *Proc. Natl. Acad. Sci. USA* 93: 14355-14360.

CHROMOSOMAL LOCATION

Genetic locus: Etfb (mouse) mapping to 7 B4.

PRODUCT

ETFB siRNA (m) is a pool of 2 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 ETFB shRNA Plasmid (m): sc-144954-SH and ETFB shRNA (m) Lentiviral Particles: sc-144954-V as alternate gene silencing products.

For independent verification of ETFB (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-144954A and sc-144954B.

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

ETFB siRNA (m) is recommended for the inhibition of ETFB expression in mouse 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

ETFB (F-1): sc-514807 is recommended as a control antibody for monitoring of ETFB 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 ETFB gene expression knockdown using RT-PCR Primer: ETFB (m)-PR: sc-144954-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.