

ATP5F1 siRNA (h): sc-88835

BACKGROUND

Mitochondrial ATP synthase is composed of two multi-subunit complexes that utilize an inner membrane electrochemical gradient to catalyze the synthesis of ATP during oxidative phosphorylation. The two multi-subunit complexes are designated F_1 and F_0 , the former of which comprises the soluble catalytic core and the latter of which comprises the membrane-spanning proton channel of ATP synthase. F_1 consists of five distinct subunits, designated ATP5A, ATP5B, ATP5C1, ATP5D and ATP5E, while F_0 consists of ten subunits, designated ATP5H, ATP5G1, ATP5I, ATP5G2, ATP5J2, ATP5J, ATP5G3, ATP5S, ATP5F1 and ATP5L. ATP5F1 (ATP synthase, H^+ transporting, mitochondrial F_0 complex, subunit B_1), also known as PIG47, is a 256 amino acid protein that localizes to the mitochondrial membrane and exists as a subunit of the F_0 complex. ATP5F1 is encoded by a gene located on human chromosome 1, which spans about 260 million base pairs and comprises nearly 8% of the human genome.

REFERENCES

1. Higuti, T., et al. 1991. Molecular cloning of cDNA for the import precursor of human coupling factor 6 of H^+ -ATP synthase in mitochondria. *Biochem. Biophys. Res. Commun.* 178: 793-799.
2. Javed, A.A., et al. 1991. Human mitochondrial ATP synthase: cloning cDNA for the nuclear-encoded precursor of coupling factor 6. *Gene* 97: 307-310.
3. Yan, W.L., et al. 1994. Sequence analysis and mapping of a novel human mitochondrial ATP synthase subunit 9 cDNA (ATP5G3). *Genomics* 24: 375-377.
4. Elston, T., et al. 1998. Energy transduction in ATP synthase. *Nature* 391: 510-513.
5. Wang, H., et al. 1998. Energy transduction in the F_1 motor of ATP synthase. *Nature* 396: 279-282.

CHROMOSOMAL LOCATION

Genetic locus: ATP5F1 (human) mapping to 1p13.2.

PRODUCT

ATP5F1 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 ATP5F1 shRNA Plasmid (h): sc-88835-SH and ATP5F1 shRNA (h) Lentiviral Particles: sc-88835-V as alternate gene silencing products.

For independent verification of ATP5F1 (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-88835A, sc-88835B and sc-88835C.

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

ATP5F1 siRNA (h) is recommended for the inhibition of ATP5F1 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

ATP5F1 (C-12): sc-514419 is recommended as a control antibody for monitoring of ATP5F1 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 ATP5F1 gene expression knockdown using RT-PCR Primer: ATP5F1 (h)-PR: sc-88835-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

RESEARCH USE

For research use only, not for use in diagnostic procedures.