SANTA CRUZ BIOTECHNOLOGY, INC.

ATP5B siRNA (h): sc-40565

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. ATP5B, also designated ATPMB, ATPSB or mitochondrial ATP synthetase, β subunit, is a 529 amino acid protein that localizes to the mitochondrial membrane and exists as a subunit of the F_0 complex. ATP5B is encoded by a nuclear gene and assembled with the other subunits encoded by both mitochondrial and nuclear genes. The ATP5B gene is activated by members of the Ets family of transcription factors, suggesting that Ets transcription factors are involved in the enhanced expression of the ATP5B gene in highly proliferating cells and in the coordinate transcription of nuclear genes for mitochondrial proteins. ATP5B mRNA levels vary among species through transcriptional control with high expression levels in heart, lower levels in skeletal muscle and the lowest levels in liver and kidney.

REFERENCES

1. Ohta, S. and Kagawa, Y. 1986. Human F_1-ATPase: molecular cloning of cDNA for the β subunit. J. Biochem. 99: 135-141.
2. Neckelmann, N., et al. 1989. The human ATP synthase β subunit gene: sequence analysis, chromosome assignment, and differential expression. Genomics 5: 829-843.

CHROMOSOMAL LOCATION

Genetic locus: ATP5B (human) mapping to 12q13.3.

PRODUCT

ATP5B 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ATP5B shRNA Plasmid (h): sc-40565-SH and ATP5B shRNA (h) Lentiviral Particles: sc-40565-V as alternate gene silencing products.

For independent verification of ATP5B (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-40565A, sc-40565B and sc-40565C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

ATP5B siRNA (h) is recommended for the inhibition of ATP5B 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 µM in 66 µl. 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

ATP5B (E-1): sc-55597 is recommended as a control antibody for monitoring of ATP5B 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 ATP5B gene expression knockdown using RT-PCR Primer: ATP5B (h)-PR: sc-40565-PR (20 µl, 469 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Guo, H., et al. 2011. Anthocyanin inhibits high glucose-induced hepatic mtGPAT1 activation and prevents fatty acid synthesis through PKCζ. J. Lipid Res. 52: 908-922.
2. Xiao, X., et al. 2013. Deregulation of mitochondrial ATPsyn-β in acute myeloid leukemia cells and with increased drug resistance. PLoS ONE 8: e83610.
3. Adachi, K., et al. 2022. A PCR-amplified transgene fragment flanked by a single copy of a truncated inverted terminal repeat for recombinant adeno-associated virus production prevents unnecessary plasmid DNA packaging. Gene Ther. 29: 449-457.

RESEARCH USE

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