SANTA CRUZ BIOTECHNOLOGY, INC.

NDUFS2 siRNA (m): sc-106290

BACKGROUND

Located in the mitochondrial inner membrane, mitochondrial complex I is the first and largest enzyme in the electron transport chain of oxidative phosphorylation. By oxidizing NADH that is produced in the Krebs cycle, this complex utilizes the two electrons to reduce ubiquinone to ubiquinol, thereby initiating the passage of electrons to successive complexes and ultimately leading to the reduction of oxygen to water. Mitochondrial complex I consists of over 40 subunits and is of considerable clinical interest since defects in any of the subunits can lead to various myopathies and neuropathies. As a subunit of mitochondrial complex I, NDUFS2 (NADH dehydrogenase [ubiquinone] iron-sulfur protein 2), also designated NADH-ubiquinone oxidoreductase 49 kDa subunit, is a 463 amino acid protein that is suggested to be required for catalytic activity. Defects in the gene encoding NDUFS2 are the cause of complex I mitochondrial respiratory chain deficiency, which is characterized by a variety of symptoms including liver failure, cardiomyopathy and neurodegeneration.

REFERENCES

1. Procaccio, V., et al. 1998. Mapping to 1q23 of the human gene (NDUFS2) encoding the 49-kDa subunit of the mitochondrial respiratory complex I and immunodetection of the mature protein in mitochondria. Mamm. Genome 9: 482-484.
2. Smeitink, J. and van den Heuvel, L. 1999. Human mitochondrial complex I in health and disease. Am. J. Hum. Genet. 64: 1505-1510.
3. Loeffen, J., et al. 2001. Mutations in the complex I NDUFS2 gene of patients with cardiomyopathy and encephalomyopathy. Ann. Neurol. 49: 195-201.
4. Bugiani, M., et al. 2004. Clinical and molecular findings in children with complex I deficiency. Biochim. Biophys. Acta 1659: 136-147.
5. Ugalde, C., et al. 2004. Differences in assembly or stability of complex I and other mitochondrial OXPHOS complexes in inherited complex I deficiency. Hum. Mol. Genet. 13: 659-667.
6. Visch, H.J., et al. 2004. Inhibition of mitochondrial Na^+-Ca^{2+} exchange restores agonist-induced ATP production and Ca^{2+} handling in human complex I deficiency. J. Biol. Chem. 279: 40328-40336.

CHROMOSOMAL LOCATION

Genetic locus: Ndufs2 (mouse) mapping to 1 H3.

PRODUCT

NDUFS2 siRNA (m) 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 NDUFS2 shRNA Plasmid (m): sc-106290-SH and NDUFS2 shRNA (m) Lentiviral Particles: sc-106290-V as alternate gene silencing products.

For independent verification of NDUFS2 (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-106290A, sc-106290B and sc-106290C.

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

NDUFS2 siRNA (m) is recommended for the inhibition of NDUFS2 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 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

NDUFS2 (B-3): sc-390596 is recommended as a control antibody for monitoring of NDUFS2 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 NDUFS2 gene expression knockdown using RT-PCR Primer: NDUFS2 (m)-PR: sc-106290-PR (20 µl, 566 bp). 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.