

NDUFS5 siRNA (h): sc-88473

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, NDUFS5 (NADH dehydrogenase [ubiquinone] iron-sulfur protein 5), also designated NADH-ubiquinone oxidoreductase 15 kDa subunit, is a 106 amino acid protein that is suggested to not be involved in catalysis. NDUFS5 is expressed ubiquitously in human tissues, with a relative higher expression in human heart, skeletal muscle, liver, kidney and fetal heart.

REFERENCES

1. Smeitink, J. and van den Heuvel, L. 1999. Human mitochondrial complex I in health and disease. *Am. J. Hum. Genet.* 64: 1505-1510.
2. Loeffen, J., et al. 1999. The human NADH: ubiquinone oxidoreductase NDUFS5 (15 kDa) subunit: cDNA cloning, chromosomal localization, tissue distribution and the absence of mutations in isolated complex I-deficient patients. *J. Inherit. Metab. Dis.* 22: 19-28.
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: NDUFS5 (human) mapping to 1p34.3.

PRODUCT

NDUFS5 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 NDUFS5 shRNA Plasmid (h): sc-88473-SH and NDUFS5 shRNA (h) Lentiviral Particles: sc-88473-V as alternate gene silencing products.

For independent verification of NDUFS5 (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-88473A, sc-88473B and sc-88473C.

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

NDUFS5 siRNA (h) is recommended for the inhibition of NDUFS5 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor NDUFS5 gene expression knockdown using RT-PCR Primer: NDUFS5 (h)-PR: sc-88473-PR (20 μl). 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.