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SANTA CRUZ BIOTECHNOLOGY, INC.

NDUFS8 siRNA (m): sc-149890

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 during the Krebs cycle, this complex utilizes 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, NDUFS8 (NADH dehydrogenase [ubiquinone] iron-sulfur protein 8), also known as TYKY, CI-23k, CI23KD or NADH-ubiquinone oxidoreductase 23 kDa subunit, is a 210 amino acid protein that is suggested to be required for catalytic activity. Defects in the gene encoding NDUFS8 are the cause of Leigh syndrome, a severe neurological disorder that is characterized by bilaterally symmetrical necrotic lesions in subcortical brain regions.

REFERENCES

1. Hyslop, S.J., et al. 1996. Assignment of the PSST subunit gene of human mitochondrial complex I to chromosome 19p13. Genomics 37: 375-380.
2. Procaccio, V., et al. 1997. cDNA sequence and chromosomal localization of the NDUFS8 human gene coding for the 23 kDa subunit of the mitochondrial complex I. Biochim. Biophys. Acta 1351: 37-41.
3. Loeffen, J., et al. 1998. The first nuclear-encoded complex I mutation in a patient with Leigh syndrome. Am. J. Hum. Genet. 63: 1598-1608.
4. de Sury, R., et al. 1998. Genomic structure of the human NDUFS8 gene coding for the iron-sulfur TYKY subunit of the mitochondrial NADH:ubiquinone oxidoreductase. Gene 215: 1-10.
5. Triepels, R., et al. 1998. The nuclear-encoded human NADH:ubiquinone oxidoreductase NDUFA8 subunit: cDNA cloning, chromosomal localization, tissue distribution, and mutation detection in complex-I-deficient patients. Hum. Genet. 103: 557-563.

CHROMOSOMAL LOCATION

Genetic locus: Ndufs8 (mouse) mapping to 19 A.

PRODUCT

NDUFS8 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 NDUFS8 shRNA Plasmid (m): sc-149890-SH and NDUFS8 shRNA (m) Lentiviral Particles: sc-149890-V as alternate gene silencing products.

For independent verification of NDUFS8 (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-149890A, sc-149890B and sc-149890C.

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 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

NDUFS8 siRNA (m) is recommended for the inhibition of NDUFS8 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

NDUFS8 (A-6): sc-515527 is recommended as a control antibody for monitoring of NDUFS8 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 NDUFS8 gene expression knockdown using RT-PCR Primer: NDUFS8 (m)-PR: sc-149890-PR (20 µl). 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.