

NDUFS4 siRNA (m): sc-75893

BACKGROUND

Complex I (also known as NADH dehydrogenase) of the electron transport chain (respiratory chain) is an enzymatic complex that catalyzes the transfer of electrons from NADH to ubiquinone. Free energy from the reaction is conserved in the transfer of protons into the intermembrane space to create an electrochemical proton gradient, a driving force for ATP synthesis. Complex I is a complicated, multi-protein, L-shaped complex composed of at least 45 different subunits and located in the mitochondrial inner membrane. NDUFS4 (NADH dehydrogenase (ubiquinone) Fe-S protein 4), also known as AQDQ or CI-18 (complex I-18 kDa protein), belongs to the complex I NDUFS4 subunit family. NDUFS4 localizes to the matrix side of the inner membrane of the mitochondrion and functions as an accessory subunit of complex I. Mutations in the gene encoding NDUFS4 can result in complex I mitochondrial respiratory chain deficiency. Patients with this deficiency may exhibit cardiomyopathy, myopathy, liver failure and neurological disorders.

REFERENCES

1. Papa, S., et al. 1996. The nuclear-encoded 18 kDa (IP) AQDQ subunit of bovine heart complex I is phosphorylated by the mitochondrial cAMP-dependent protein kinase. *FEBS Lett.* 379: 299-301.
2. Online Mendelian Inheritance in Man, OMIM™. 1998. Johns Hopkins University, Baltimore, MD. MIM Number: 602694. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Budde, S.M., et al. 2000. Combined enzymatic complex I and III deficiency associated with mutations in the nuclear encoded NDUFS4 gene. *Biochem. Biophys. Res. Commun.* 275: 63-68.
4. Scacco, S., et al. 2006. Mutations in structural genes of complex I associated with neurological diseases. *Ital. J. Biochem.* 55: 254-262.
5. Piccoli, C., et al. 2006. cAMP controls oxygen metabolism in mammalian cells. *FEBS Lett.* 580: 4539-4543.
6. Iuso, A., et al. 2006. Dysfunctions of cellular oxidative metabolism in patients with mutations in the NDUFS1 and NDUFS4 genes of complex I. *J. Biol. Chem.* 281: 10374-10380.

CHROMOSOMAL LOCATION

Genetic locus: *Ndusf4* (mouse) mapping to 13 D2.2.

PRODUCT

NDUFS4 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NDUFS4 shRNA Plasmid (m): sc-75893-SH and NDUFS4 shRNA (m) Lentiviral Particles: sc-75893-V as alternate gene silencing products.

For independent verification of NDUFS4 (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-75893A, sc-75893B and sc-75893C.

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

NDUFS4 siRNA (m) is recommended for the inhibition of NDUFS4 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 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

NDUFS4 (1-E-4): sc-100567 is recommended as a control antibody for monitoring of NDUFS4 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 NDUFS4 gene expression knockdown using RT-PCR Primer: NDUFS4 (m)-PR: sc-75893-PR (20 μ l). 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.