

NUBPL siRNA (h): sc-92266

BACKGROUND

NUBPL (nucleotide-binding protein-like) is a 319 amino acid mitochondrial protein that belongs to the Mrp/NBP35 ATP-binding protein family and exists as two alternatively spliced isoforms. Required for the assembly of the mitochondrial membrane respiratory chain NADH dehydrogenase (complex I), NUBPL may deliver one or more Fe-S clusters to complex I subunits. With highest expression in liver and kidney, NUBPL is expressed at significant levels in small intestine and brain. Defects in NUBPL are a cause of mitochondrial complex I deficiency (MT-C1D), a disorder of the mitochondrial respiratory chain that causes a wide range of afflictions from lethal neonatal disease to adult-onset neurodegenerative disorders. Phenotypes include macrocephaly with progressive leukodystrophy, non-specific encephalopathy, cardiomyopathy, myopathy, liver disease, Leigh syndrome, Leber hereditary optic neuropathy and some forms of Parkinson disease.

REFERENCES

1. Online Mendelian Inheritance in Man, OMIM™. 1987. Johns Hopkins University, Baltimore, MD. MIM Number: 252010. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
2. Larsson, N.G., et al. 1991. Leber's hereditary optic neuropathy and complex I deficiency in muscle. *Ann. Neurol.* 30: 701-708.
3. Robinson, B.H. 1998. Human complex I deficiency: clinical spectrum and involvement of oxygen free radicals in the pathogenicity of the defect. *Biochim. Biophys. Acta* 1364: 271-286.
4. Smeitink, J., et al. 1999. Human mitochondrial complex I in health and disease. *Am. J. Hum. Genet.* 64: 1505-1510.
5. Triepels, R.H., et al. 2001. Respiratory chain complex I deficiency. *Am. J. Med. Genet.* 106: 37-45.
6. Tretter, L., et al. 2004. Initiation of neuronal damage by complex I deficiency and oxidative stress in Parkinson's disease. *Neurochem. Res.* 29: 569-577.
7. Sheftel, A.D., et al. 2009. Human ind1, an iron-sulfur cluster assembly factor for respiratory complex I. *Mol. Cell. Biol.* 29: 6059-6073.

CHROMOSOMAL LOCATION

Genetic locus: NUBPL (human) mapping to 14q12.

PRODUCT

NUBPL 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see NUBPL shRNA Plasmid (h): sc-92266-SH and NUBPL shRNA (h) Lentiviral Particles: sc-92266-V as alternate gene silencing products.

For independent verification of NUBPL (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-92266A, sc-92266B and sc-92266C.

RESEARCH USE

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

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

NUBPL siRNA (h) is recommended for the inhibition of NUBPL 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 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

NUBPL (G-11): sc-398217 is recommended as a control antibody for monitoring of NUBPL 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 NUBPL gene expression knockdown using RT-PCR Primer: NUBPL (h)-PR: sc-92266-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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