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COX15 siRNA (m): sc-142524

BACKGROUND

The cytochrome c oxidase (COX) family of proteins function as the final electron donor in the respiratory chain to drive a proton gradient across the inner mitochondrial membrane, ultimately resulting in the production of water. The mammalian COX apoenzyme is a dimer, with each monomer consisting of 13 subunits, some of which are mitochondrial and some of which are nuclear. COX15 (cytochrome c oxidase assembly protein COX15 homolog) is a 410 amino acid multi-pass mitochondrial membrane protein belonging to the COX15/ctaA family. Existing as two alternatively spliced isoforms, COX15 is predominantly expressed in tissues containing high rates of oxidative phosphorylation including brain, heart and muscle. COX15 defects are the cause of a neurodegenerative disorder known as Leigh syndrome and cytochrome c oxidase deficiency (COX deficiency).

REFERENCES

1. Kennaway, N.G., et al. 1990. Isoforms of mammalian cytochrome c oxidase: correlation with human cytochrome c oxidase deficiency. *Pediatr. Res.* 28: 529-535.
2. Petruzzella, V., et al. 1998. Identification and characterization of human cDNAs specific to BCS1, PET112, SCO1, COX15, and COX11, five genes involved in the formation and function of the mitochondrial respiratory chain. *Genomics* 54: 494-504.
3. Barros, M.H., et al. 2001. Involvement of mitochondrial ferredoxin and Cox15p in hydroxylation of heme O. *FEBS Lett.* 492: 133-138.
4. Antonicka, H., et al. 2003. Mutations in COX15 produce a defect in the mitochondrial heme biosynthetic pathway, causing early-onset fatal hypertrophic cardiomyopathy. *Am. J. Hum. Genet.* 72: 101-114.
5. Oquendo, C.E., et al. 2004. Functional and genetic studies demonstrate that mutation in the COX15 gene can cause Leigh syndrome. *J. Med. Genet.* 41: 540-544.
6. Vitali, M., et al. 2009. Analysis of the genes coding for subunit 10 and 15 of cytochrome c oxidase in Alzheimer's disease. *J. Neural. Transm.* 116: 1635-1641.

CHROMOSOMAL LOCATION

Genetic locus: Cox15 (mouse) mapping to 19 C3.

PRODUCT

COX15 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 COX15 shRNA Plasmid (m): sc-142524-SH and COX15 shRNA (m) Lentiviral Particles: sc-142524-V as alternate gene silencing products.

For independent verification of COX15 (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-142524A, sc-142524B and sc-142524C.

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

COX15 siRNA (m) is recommended for the inhibition of COX15 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

COX15 (G-10): sc-390987 is recommended as a control antibody for monitoring of COX15 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 COX15 gene expression knockdown using RT-PCR Primer: COX15 (m)-PR: sc-142524-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.