

Mfn1 siRNA (r): sc-270320

BACKGROUND

Mitofusin 1 (Mfn1) and mitofusin 2 (Mfn2) are homologs for the *Drosophila* protein fuzzy onion (Fzo). They are mitochondrial membrane proteins and are mediators of mitochondrial fusion. A GTPase domain is required for Mfn protein function but the molecular mechanisms of the GTPase-dependent reaction as well as the functional division of the two Mfn proteins are unknown. They are essential for embryonic development and may play a role in the pathobiology of obesity. Although the Mfn1 and Mfn2 genes are broadly expressed, they show different levels of expression in different tissues. Two Mfn1 transcripts are elevated in heart, while Mfn2 mRNA is abundantly expressed in heart and muscle tissue but present only at low levels in many other tissues. Mfn1 localizes to mitochondria and participates in at least two different high molecular weight protein complexes in a GTP-dependent manner. Purified recombinant Mfn1 exhibited approximately eightfold higher GTPase activity than Mfn2.

REFERENCES

1. Santel, A., et al. 2001. Control of mitochondrial morphology by a human mitofusin. *J. Cell Sci.* 114: 867-874.
2. Rojo, M., et al. 2002. Membrane topology and mitochondrial targeting of mitofusins, ubiquitous mammalian homologs of the transmembrane GTPase Fzo. *J. Cell Sci.* 115: 1663-1674.
3. Chen, H., et al. 2003. Mitofusins Mfn1 and Mfn2 coordinately regulate mitochondrial fusion and are essential for embryonic development. *J. Cell Biol.* 160: 189-200.
4. Bach, D., et al. 2003. Mitofusin-2 determines mitochondrial network architecture and mitochondrial metabolism a novel regulatory mechanism altered in obesity. *J. Biol. Chem.* 278: 17190-17197.
5. Santel, A., et al. 2003. Mitofusin-1 protein is a generally expressed mediator of mitochondrial fusion in mammalian cells. *J. Cell Sci.* 116: 2763-2774.
6. Ishihara, N., et al. 2004. Mitofusin 1 and 2 play distinct roles in mitochondrial fusion reactions via GTPase activity. *J. Cell Sci.* 117: 6535-6546.

CHROMOSOMAL LOCATION

Genetic locus: Mfn1 (rat) mapping to 2q24.

PRODUCT

Mfn1 siRNA (r) 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 Mfn1 shRNA Plasmid (r): sc-270320-SH and Mfn1 shRNA (r) Lentiviral Particles: sc-270320-V as alternate gene silencing products.

For independent verification of Mfn1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270320A, sc-270320B and sc-270320C.

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

Mfn1 siRNA (r) is recommended for the inhibition of Mfn1 expression in rat 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

Mfn1 (D-10): sc-166644 is recommended as a control antibody for monitoring of Mfn1 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 Mfn1 gene expression knockdown using RT-PCR Primer: Mfn1 (r)-PR: sc-270320-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.