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MFF siRNA (m): sc-149404

BACKGROUND

MFF (mitochondrial fission factor), also known as GL004, AD030 or AD033, is a 342 amino acid single-pass type IV membrane protein of the mitochondrial outer membrane that belongs to the tango11 family. Involved in mitochondrial and peroxisomal fission, MFF is abundantly expressed in stomach, heart, muscle, liver, brain and kidney. MFF exists as five alternatively spliced isoforms that are encoded by a gene that maps to human chromosome 2q36.3. As the second largest human chromosome, chromosome 2 consists of 237 million bases, encodes over 1,400 genes and makes up approximately 8% of the human genome. A number of genetic diseases are linked to genes on chromosome 2. Harlequin ichthyosis, a rare and morbid skin deformity, is associated with mutations in the ABCA12 gene. The lipid metabolic disorder sitosterolemia is associated with ABCG5 and ABCG8. An extremely rare recessive genetic disorder, Alström syndrome, is due to mutations in the ALMS1 gene.

REFERENCES

1. Patel, S.B., et al. 1998. Mapping a gene involved in regulating dietary cholesterol absorption. The sitosterolemia locus is found at chromosome 2p21. *J. Clin. Invest.* 102: 1041-1044.
2. Zumsteg, U., et al. 2000. Alstrom syndrome: confirmation of linkage to chromosome 2p12-13 and phenotypic heterogeneity in three affected sibs. *J. Med. Genet.* 37: E8.
3. Shulenin, S., et al. 2001. An ATP-binding cassette gene (ABCG5) from the ABCG (White) gene subfamily maps to human chromosome 2p21 in the region of the Sitosterolemia locus. *Cytogenet. Cell Genet.* 92: 204-208.
4. Hearn, T., et al. 2002. Mutation of ALMS1, a large gene with a tandem repeat encoding 47 amino acids, causes Alström syndrome. *Nat. Genet.* 31: 79-83.
5. Kelsell, D.P., et al. 2005. Mutations in ABCA12 underlie the severe congenital skin disease harlequin ichthyosis. *Am. J. Hum. Genet.* 76: 794-803.
6. Gandre-Babbe, S., et al. 2008. The novel tail-anchored membrane protein Mff controls mitochondrial and peroxisomal fission in mammalian cells. *Mol. Biol. Cell* 19: 2402-2412.

CHROMOSOMAL LOCATION

Genetic locus: Mff (mouse) mapping to 1 C5.

PRODUCT

MFF siRNA (m) is a pool of 2 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 MFF shRNA Plasmid (m): sc-149404-SH and MFF shRNA (m) Lentiviral Particles: sc-149404-V as alternate gene silencing products.

For independent verification of MFF (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-149404A and sc-149404B.

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

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

MFF (B-2): sc-398617 is recommended as a control antibody for monitoring of MFF 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 MFF gene expression knockdown using RT-PCR Primer: MFF (m)-PR: sc-149404-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.