SANTA CRUZ BIOTECHNOLOGY, INC.

Mpi siRNA (h): sc-90211

BACKGROUND

Mpi (mannose phosphate isomerase), also known as PMI (phosphomannose isomerase) or PMI1, is a 423 amino acid zinc metalloenzyme belonging to the mannose-6-phosphate isomerase type 1 family, and is expressed in all tissues, more abundantly in heart, brain and skeletal muscle. A steady supply of D-mannose derivatives, which are required for most glycosylation reactions, is maintained by Mpi. Localized to the cytoplasm, Mpi utilizes zinc as a cofactor and catalyzes the interconversion of fructose-6-phosphate and mannose-6-phosphate. Mutations in the gene encoding Mpi lead to congenital disorder of glycosylation type 1B (CDG1B), also designated carbohydrate-deficient glycoprotein syndrome type Ib (CDGS1B), which is characterized by protein-losing enteropathy. Congenital disorders of glycosylation are metabolic deficiencies in glycoprotein biosynthesis that usually results in severe mental and psychomotor retardation.

REFERENCES

1. Proudfoot, A.E., et al. 1996. *In vivo* and *in vitro* folding of a recombinant metalloenzyme, phosphomannose isomerase. Biochem. J. 318: 437-442.
2. Jaeken, J., et al. 1998. Phosphomannose isomerase deficiency: a carbohydrate-deficient glycoprotein syndrome with hepatic-intestinal presentation. Am. J. Hum. Genet. 62: 1535-1539.
3. Niehues, R., et al. 1998. Carbohydrate-deficient glycoprotein syndrome type Ib. Phosphomannose isomerase deficiency and mannose therapy. J. Clin. Invest. 101: 1414-1420.
4. Schollen, E., et al. 2000. Genomic organization of the human phosphomannose isomerase (MPI) gene and mutation analysis in patients with congenital disorders of glycosylation type Ib (CDG-Ib). Hum. Mutat. 16: 247-252.
5. de Lonlay, P., et al. 2001. A broad spectrum of clinical presentations in congenital disorders of glycosylation I: a series of 26 cases. J. Med. Genet. 38: 14-19.
6. Schollen, E., et al. 2002. DHPLC analysis as a platform for molecular diagnosis of congenital disorders of glycosylation (CDG). Eur. J. Hum. Genet. 10: 643-648.

CHROMOSOMAL LOCATION

Genetic locus: MPI (human) mapping to 15q24.1.

PRODUCT

Mpi 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Mpi shRNA Plasmid (h): sc-90211-SH and Mpi shRNA (h) Lentiviral Particles: sc-90211-V as alternate gene silencing products.

For independent verification of Mpi (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-90211A, sc-90211B and sc-90211C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Mpi siRNA (h) is recommended for the inhibition of Mpi 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 µM in 66 µl. 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

Mpi (B-2): sc-393484 is recommended as a control antibody for monitoring of Mpi 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Mpi gene expression knockdown using RT-PCR Primer: Mpi (h)-PR: sc-90211-PR (20 µl). 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.