

MAN1B1 siRNA (h): sc-92479

BACKGROUND

MAN1B1 (mannosidase α , class 1B, member 1), also referred to as MANA-ER or ERManI, is a widely expressed enzyme that is a member of the glycosyl hydrolase 47 family. MAN1B1 is a single-pass type II membrane protein that localizes to the endoplasmic reticulum (ER) and catalyzes the first mannosyl trimming step in the maturation of Asn-linked oligosaccharide biosynthesis on glycoproteins. Asn-linked oligosaccharides are important for a variety of biological functions, including cellular recognition, adhesion and protein targeting. MAN1B1 is also involved in targeting terminally misfolded or unassembled glycoproteins for degradation via the cytoplasmic ubiquitin-proteasome pathway, a process known as endoplasmic reticulum-associated protein degradation (ERAD). MAN1B1 activity requires calcium and is inhibited by either 1-deoxymannojirimycin or kifunensine, which are class I α -mannosidase inhibitors.

REFERENCES

1. Tremblay, L.O. and Herscovics, A. 1999. Cloning and expression of a specific human α 1,2-mannosidase that trims Man₅GlcNAc₂ to Man₆GlcNAc₂ isomer B during N-glycan biosynthesis. *Glycobiology* 9: 1073-1078.
2. Gonzalez, D.S., et al. 1999. Identification, expression, and characterization of a cDNA encoding human endoplasmic reticulum mannosidase I, the enzyme that catalyzes the first mannosyl trimming step in mammalian Asn-linked oligosaccharide biosynthesis. *J. Biol. Chem.* 274: 21375-21386.
3. Online Mendelian Inheritance in Man, OMIM[™]. 1999. Johns Hopkins University, Baltimore, MD. MIM Number: 604346. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Hosokawa, N., et al. 2003. Enhancement of endoplasmic reticulum (ER) degradation of misfolded Null Hong Kong α 1-antitrypsin by human ER mannosidase I. *J. Biol. Chem.* 278: 26287-26294.

CHROMOSOMAL LOCATION

Genetic locus: MAN1B1 (human) mapping to 9q34.3.

PRODUCT

MAN1B1 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 MAN1B1 shRNA Plasmid (h): sc-92479-SH and MAN1B1 shRNA (h) Lentiviral Particles: sc-92479-V as alternate gene silencing products.

For independent verification of MAN1B1 (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-92479A, sc-92479B and sc-92479C.

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.

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

MAN1B1 siRNA (h) is recommended for the inhibition of MAN1B1 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

MAN1B1 (E-10): sc-393145 is recommended as a control antibody for monitoring of MAN1B1 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 MAN1B1 gene expression knockdown using RT-PCR Primer: MAN1B1 (h)-PR: sc-92479-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.