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CD-MPR shRNA (m) Lentiviral Particles: sc-45451-V

BACKGROUND

CD-MPR (cation-dependent mannose-6-phosphate receptor) is an oligomeric transmembrane protein that plays a critical role in the intracellular delivery of phosphorylated lysosomal enzymes from the *trans*-Golgi network (TGN). Intracellular trafficking of CD-MPR is mediated by sorting signals in its 67 amino acid cytoplasmic tail, which prevent it from entering the lysosome, where it would be degraded. CD-MPR is predominantly expressed in mouse testicular germ cells and shows differentiated expression during maturation of rat spermatozoa. Increased expression of CD-MPR in Alzheimer's disease and the location of the CD-MPR gene next to a region on chromosome 12 which is possibly linked to the disease indicate that CD-MPR may play a role in Alzheimer's disease.

REFERENCES

1. Sleat, D.E., et al. 1997. Ligand binding specificities of the two mannose 6-phosphate receptors. *J. Biol. Chem.* 272: 731-738.
2. Schweizer, A., et al. 1997. Proper sorting of the cation-dependent mannose 6-phosphate receptor in endosomes depends on a pair of aromatic amino acids in its cytoplasmic tail. *Proc. Natl. Acad. Sci. USA* 94: 14471-14476.
3. Olson, L.J., et al. 1999. Mutational analysis of the binding site residues of the bovine cation-dependent mannose 6-phosphate receptor. *J. Biol. Chem.* 274: 36905-36911.
4. Chayko, C.A., et al. 2000. Targeted disruption of the cation-dependent or cation-independent mannose 6-phosphate receptor does not decrease the content of acid glycosidases in the acrosome. *J. Androl.* 21: 944-953.
5. Belmonte, S.A., et al. 2000. Changes in distribution of phosphomannosyl receptors during maturation of rat spermatozoa. *Biol. Reprod.* 63: 1172-1178.

CHROMOSOMAL LOCATION

Genetic locus: M6pr (mouse) mapping to 6 F1.

PRODUCT

CD-MPR shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see CD-MPR siRNA (m): sc-45451 and CD-MPR shRNA Plasmid (m): sc-45451-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80°C . Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4°C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

APPLICATIONS

CD-MPR shRNA (m) Lentiviral Particles is recommended for the inhibition of CD-MPR expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

CD-MPR (H-251): sc-66826 is recommended as a control antibody for monitoring of CD-MPR 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker[™] compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker[™] Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz[™] Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor CD-MPR gene expression knockdown using RT-PCR Primer: CD-MPR (m)-PR: sc-45451-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.