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Cyclophilin 40 shRNA (h) Lentiviral Particles: sc-44892-V

BACKGROUND

Cyclophilins are conserved, ubiquitous and abundant cytosolic peptidylprolyl *cis-trans* isomerases that accelerate the isomerization of XaaPro peptide bonds and the refolding of proteins. Human cyclophilin A (CyPA), an intracellular protein of 165 amino acids, is the target of cyclosporin A (CsA) and is encoded by a single unique gene conserved from yeast to humans. Cyclophilin B (CyPB) is secreted in biological fluids such as blood or milk and binds to a specific receptor present on the human lymphoblastic cell line Jurkat and on human peripheral blood lymphocytes. Cyclophilin 40 (CyP40) is a widely expressed cytoplasmic protein that catalyzes the *cis-trans* isomerization of proline imidic peptide bonds in oligopeptides. It is a widely expressed cytoplasmic protein.

REFERENCES

1. Kieffer, L.J., et al. 1993. Cyclophilin-40, a protein with homology to the P59 component of the steroid receptor complex. Cloning of the cDNA and further characterization. *J. Biol. Chem.* 268: 12303-12310.
2. Yokoi, H., et al. 1996. The structure and complete nucleotide sequence of the human Cyclophilin 40 (PPID) gene. *Genomics* 35: 448-455.
3. Gevaert, K., et al. 2003. Exploring proteomes and analyzing protein processing by mass spectrometric identification of sorted N-terminal peptides. *Nat. Biotechnol.* 21: 566-569.
4. Guo, H.X., et al. 2005. Novel Cyclophilin D inhibitors derived from quinoxaline exhibit highly inhibitory activity against rat mitochondrial swelling and Ca²⁺ uptake/ release. *Acta Pharmacol. Sin.* 26: 1201-1211.
5. Schinzel, A.C., et al. 2005. Cyclophilin D is a component of mitochondrial permeability transition and mediates neuronal cell death after focal cerebral ischemia. *Proc. Natl. Acad. Sci. USA* 102: 12005-12010.

CHROMOSOMAL LOCATION

Genetic locus: PPID (human) mapping to 4q32.1.

PRODUCT

Cyclophilin 40 shRNA (h) 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 Cyclophilin 40 siRNA (h): sc-44892 and Cyclophilin 40 shRNA Plasmid (h): sc-44892-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

Cyclophilin 40 siRNA (h) is recommended for the inhibition of Cyclophilin 40 expression in human 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

Cyclophilin 40 (F-5): sc-137157 is recommended as a control antibody for monitoring of Cyclophilin 40 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-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Cyclophilin 40 gene expression knockdown using RT-PCR Primer: Cyclophilin 40 (h)-PR: sc-44892-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° 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.