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LRP5 shRNA (h) Lentiviral Particles: sc-43900-V

BACKGROUND

Members of the LDL receptor gene family, including LDLR (low density lipoprotein receptor), LRP5 (low density lipoprotein related proteins), Megalin (also designated GP330), VLDLR (very low density lipoprotein receptor) and ApoER2, are characterized by a cluster of cysteine-rich class A repeats, epidermal growth factor (EGF)-like repeats, YWTD repeats and an O-linked sugar domain. Of the known family members, LRP5 is most closely related to LRP1. However, LRP5 has a unique organization of EGF and LDLR repeats compared to other LDLR family members and likely represents a new category in this family. LRP5 is expressed in rat tibia in areas of the bone that are involved in remodeling. LRP5 is a Wnt coreceptor that binds to Axin and regulates the canonical Wnt signaling pathway. LRP5 affects bone mass accrual during growth and mutations in LRP5 cause the autosomal recessive disorder osteoporosis-pseudoglioma syndrome (OPPG). The gene which encodes LRP5 maps to human chromosome 11q13.2.

REFERENCES

- Hey, P.J., et al. 1998. Cloning of a novel member of the low-density lipoprotein receptor family. *Gene* 216: 103-111.
- Trommsdorff, M., et al. 1999. Reeler/Disabled-like disruption of neuronal migration in knockout mice lacking the VLDL Receptor and ApoE Receptor 2. *Cell* 97: 689-701.
- Mikhailenko, I., et al. 1999. Functional domains of the very low density lipoprotein receptor: molecular analysis of ligand binding and acid-dependent ligand dissociation mechanisms. *J. Cell Sci.* 112: 3269-3281.
- Chen, D., et al. 1999. Molecular cloning of mouse Lrp7(Lr3) cDNA and chromosomal mapping of orthologous genes in mouse and human. *Genomics* 55: 314-321.

CHROMOSOMAL LOCATION

Genetic locus: LRP5 (human) mapping to 11q13.2.

PRODUCT

LRP5 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 LRP5 siRNA (h): sc-43900 and LRP5 shRNA Plasmid (h): sc-43900-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

LRP5 shRNA (h) Lentiviral Particles is recommended for the inhibition of LRP5 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

LRP5 (B-9): sc-390267 is recommended as a control antibody for monitoring of LRP5 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 LRP5 gene expression knockdown using RT-PCR Primer: LRP5 (h)-PR: sc-43900-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ 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.