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GLP-1R shRNA (m) Lentiviral Particles: sc-45764-V

BACKGROUND

Glucagon, a pancreatic hormone, functions as an antagonist to Insulin, stimulating the conversion of glycogen to glucose and increasing blood sugar levels. GLP-1 functions as a transmitter in the central nervous system, inhibiting feeding and drinking behavior. Both Glucagon and GLP-1 function through their specific binding to the Glucagon receptor or GLP-1R, respectively. The Glucagon receptor shows expression in liver, kidney and adipose tissue. GLP-1R expression primarily localizes to areas of the hypothalamus involved in feeding behavior. Both receptors and their ligands serve as potential targets for the therapeutic treatment of diabetes.

REFERENCES

- Iwanij, V., et al. 1990. Characterization of the Glucagon receptor and its functional domains using monoclonal antibodies. *J. Biol. Chem.* 265: 21302-21308.
- Rouille, Y., et al. 1995. Differential processing of proGlucagon by the subtilisin-like prohormone convertases PC2 and PC3 to generate either Glucagon or Glucagon-like peptide. *J. Biol. Chem.* 270: 26488-26496.
- Scrocchi, L.A., et al. 1996. Glucose intolerance but normal satiety in mice with a null mutation in the Glucagon-like peptide 1 receptor gene. *Nat. Med.* 2: 1254-1258.
- Bollen, M., et al. 1998. Specific features of glycogen metabolism in the liver. *Biochem. J.* 336: 19-31.
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CHROMOSOMAL LOCATION

Genetic locus: *Glp1r* (mouse) mapping to 17 A3.3.

PRODUCT

GLP-1R 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 GLP-1R siRNA (m): sc-45764 and GLP-1R shRNA Plasmid (m): sc-45764-SH as alternate gene silencing products.

APPLICATIONS

GLP-1R shRNA (m) Lentiviral Particles is recommended for the inhibition of GLP-1R expression in mouse cells.

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.

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

GLP-1R (Y-12): sc-34637 is recommended as a control antibody for monitoring of GLP-1R 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 donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GLP-1R gene expression knockdown using RT-PCR Primer: GLP-1R (m)-PR: sc-45764-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.

PROTOCOLS

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