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# PTG shRNA (h) Lentiviral Particles: sc-44047-V

## BACKGROUND

Protein phosphatase 1 (PP1) is a serine-threonine protein phosphatase that plays a central role in mediating the effects of Insulin on glucose and lipid metabolism. PTG (protein targeting to glycogen) was cloned from 3T3-L1 adipocytes as a protein that binds to the PP1 catalytic subunit. The human homolog of PTG, designated PPP1R5, has been shown to bind to PP1 and to modulate its specificity. PTEN/PPP1R5 shows 42% identity to the glycogen binding subunit, G<sub>L</sub>, of rat liver PP1. PTG is expressed predominantly in Insulin-sensitive tissues, and it localizes PP1 to glycogen. PTG also has been shown to interact with several enzymes involved in the hormonal regulation of glycogen metabolism, including phosphorylase kinase, phosphorylase A and glycogen synthase. These data indicate a role for PTG in glycogen metabolism, possibly that of a molecular scaffold.

## REFERENCES

- Cohen, P. 1989. The structure and regulation of protein phosphatases. *Annu. Rev. Biochem.* 58: 453-508.
- Saltiel, A.R. 1996. Diverse signaling pathways in the cellular actions of Insulin. *Am. J. Physiol.* 270: E375-E385.
- Doherty, M.J., et al. 1996. Amino acid sequence of a novel protein phosphatase 1 binding protein (R5) which is related to the liver- and muscle-specific glycogen binding subunits of protein phosphatase 1. *FEBS Lett.* 399: 339-343.
- Printen, J.A., et al. 1997. PTG, a protein phosphatase-1 binding protein with a role in glycogen metabolism. *Science* 275: 1475-1478.
- Armstrong, C.G., et al. 1997. PPP1R6, a novel member of the family of glycogen-targeting subunits of protein phosphatase 1. *FEBS Lett.* 418: 210-214.
- Brady, M.J., et al. 1997. Role of protein targeting to glycogen (PTG) in the regulation of protein phosphatase-1 activity. *J. Biol. Chem.* 272: 20198-20204.

## CHROMOSOMAL LOCATION

Genetic locus: PPP1R3C (human) mapping to 10q23.32.

## PRODUCT

PTG 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 µl frozen stock containing 1.0 x 10<sup>6</sup> infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suit-able for 10-20 transductions. Also see PTG siRNA (h): sc-44047 and PTG shRNA Plasmid (h): sc-44047-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.

## APPLICATIONS

PTG shRNA (h) Lentiviral Particles is recommended for the inhibition of PTG expression in human cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10<sup>6</sup> 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

PTG (N-19): sc-6582 is recommended as a control antibody for monitoring of PTG 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 PTG gene expression knockdown using RT-PCR Primer: PTG (h)-PR: sc-44047-PR (20 µl). 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.

## PROTOCOLS

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