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# DGAT2 shRNA (m) Lentiviral Particles: sc-45521-V

## BACKGROUND

Glucose and Insulin are anabolic signals which upregulate the transcriptions of a series of lipogenic enzymes to convert excess carbohydrate into triglycerides for efficient energy storage. Acyl-coenzyme A:diacylglycerol acyltransferase, also known as DGAT1 and ARGPI, is a microsomal enzyme that assists in the synthesis of fatty acids into triglycerides. DGAT1 catalyzes the terminal and only committed step in triacylglycerol synthesis by using diacylglycerol (DAG) and fatty acyl-CoA as substrates. DGAT1 plays a fundamental role in the metabolism of cellular diacylglycerol and is important in higher eukaryotes for physiologic processes involving triacylglycerol metabolism, such as intestinal fat absorption, lipoprotein assembly, adipose tissue formation and lactation. DGAT2, which has no homology to DGAT1, differs from DGAT1 in that its activity has been shown to be inhibited by MgCl in an *in vitro* assay. DGAT2 is expressed primarily in liver and white adipose tissue, which suggests that it plays an important role in mammalian triglyceride metabolism.

## REFERENCES

1. Cases, S., et al. 1998. Identification of a gene encoding an acyl-CoA:diacylglycerol acyltransferase, a key enzyme in triacylglycerol synthesis. *Proc. Natl. Acad. Sci. USA* 95: 13018-13023.
2. Cases, S., et al. 2001. Cloning of DGAT2, a second mammalian diacylglycerol acyltransferase, and related family members. *J. Biol. Chem.* 276: 38870-38876.
3. Winter, A., et al. 2002. Association of a Lysine 232/alanine polymorphism in a bovine gene encoding acyl-CoA: diacylglycerol acyltransferase (DGAT1) with variation at a quantitative trait locus for milk fat content. *Proc. Natl. Acad. Sci. USA* 99: 9300-9305.

## CHROMOSOMAL LOCATION

Genetic locus: Dgat2 (mouse) mapping to 7 E2.

## PRODUCT

DGAT2 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  $\mu$ l frozen stock containing  $1.0 \times 10^6$  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 DGAT2 siRNA (m): sc-45521 and DGAT2 shRNA Plasmid (m): sc-45521-SH as alternate gene silencing products.

## STORAGE

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

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) or our catalog for detailed protocols and support products.

## APPLICATIONS

DGAT2 shRNA (m) Lentiviral Particles is recommended for the inhibition of DGAT2 expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l frozen viral stock containing  $1.0 \times 10^6$  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

DGAT2 (H-70): sc-66859 is recommended as a control antibody for monitoring of DGAT2 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 DGAT2 gene expression knockdown using RT-PCR Primer: DGAT2 (m)-PR: sc-45521-PR (20  $\mu$ l, 544 bp). 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.