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SANTA CRUZ BIOTECHNOLOGY, INC.

DGAT2 siRNA (h): sc-45520

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 ARGP1, 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.
4. Meegalla, R.L., et al. 2002. Concerted elevation of acyl-coenzyme A:diacylglycerol acyltransferase (DGAT) activity through independent stimulation of mRNA expression of DGAT1 and DGAT2 by carbohydrate and insulin. Biochem. Biophys. Res. Commun. 298: 317-323.

CHROMOSOMAL LOCATION

Genetic locus: DGAT2 (human) mapping to 11q13.5.

PRODUCT

DGAT2 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3.3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see DGAT2 shRNA Plasmid (h): sc-45520-SH and DGAT2 shRNA (h) Lentiviral Particles: sc-45520-V as alternate gene silencing products.

For independent verification of DGAT2 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-45520A, sc-45520B and sc-45520C.

PROTOCOLS

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

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20° C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20° C, avoid contact with RNAses and repeated freeze thaw cycles.

Resuspend lyophilized siRNA duplex in 330 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

DGAT2 siRNA (h) is recommended for the inhibition of DGAT2 expression in human cells.

SUPPORT REAGENTS

For optimal siRNA transfection efficiency, Santa Cruz Biotechnology's siRNA Transfection Reagent: sc-29528 (0.3 ml), siRNA Transfection Medium: sc-36868 (20 ml) and siRNA Dilution Buffer: sc-29527 (1.5 ml) are recommended. Control siRNAs or Fluorescein Conjugated Control siRNAs are available as 10 µM in 66 µl. Each contain a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA. Fluorescein Conjugated Control siRNAs include: sc-36869, sc-44239, sc-44240 and sc-44241. Control siRNAs include: sc-37007, sc-44230, sc-44231, sc-44232, sc-44233, sc-44234, sc-44235, sc-44236, sc-44237 and sc-44238.

GENE EXPRESSION MONITORING

DGAT2 (4C1): sc-293211 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 reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz® Mounting Medium: sc-24941 or UltraCruz® Hard-set Mounting Medium: sc-359850.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DGAT2 gene expression knockdown using RT-PCR Primer: DGAT2 (h)-PR: sc-45520-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.