SANTA CRUZ BIOTECHNOLOGY, INC.

GPAM siRNA (h): sc-90652

BACKGROUND

GPAM (glycerol-3-phosphate acyltransferase, mitochondrial), also known as GPAT1, GPAT or KIAA1560, is an 828 amino acid multi-pass membrane protein that localizes to the outer membrane of the mitochondria and is involved in phospholipid metabolism. More specifically, GPAM functions to catalyze the first and committing step in the biosynthesis of glycerolipid, namely the conversion of Acyl-CoA and sn-glycerol 3-phosphate to CoA and 1-acyl-sn-glycerol 3-phosphate. Via its catalytic activity, GPAM plays an essential role in the regulation of cellular triacylglycerol and phospholipid levels. The gene encoding GPAM maps to human chromosome 10, which houses over 1,200 genes and comprises nearly 4.5% of the human genome. Defects in some of the genes that map to chromosome 10 are associated with Charcot-Marie-Tooth disease, Jackson-Weiss syndrome, Usher syndrome, nonsyndromatic deafness, Wolman's syndrome, Cowden syndrome, multiple endocrine neoplasia type 2 and porphyria.

REFERENCES

1. Skorve, J., et al. 1990. Effect of 3- and 4-thia-substituted fatty acids on glycerolipid metabolism and mitochondrial β-oxidation in rat liver. Biochem. Pharmacol. 40: 2005-2012.
2. Thomas, P.D., et al. 1990. Lipid peroxidation inactivates rat liver microsomal glycerol-3-phosphate acyl transferase. Effect of iron and copper salts and carbon tetrachloride. J. Biol. Chem. 265: 2684-2691.
3. Shin, D.H., et al. 1991. Transcriptional regulation of p90 with sequence homology to *Escherichia coli* glycerol-3-phosphate acyltransferase. J. Biol. Chem. 266: 23834-23839.
4. Yet, S.F., et al. 1993. Expression and identification of p90 as the murine mitochondrial glycerol-3-phosphate acyltransferase. Biochemistry 32: 9486-9491.
5. Jerkins, A.A., et al. 1995. Characterization of the murine mitochondrial glycerol-3-phosphate acyltransferase promoter. J. Biol. Chem. 270: 1416-1421.

CHROMOSOMAL LOCATION

Genetic locus: GPAM (human) mapping to 10q25.2.

PRODUCT

GPAM 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 GPAM shRNA Plasmid (h): sc-90652-SH and GPAM shRNA (h) Lentiviral Particles: sc-90652-V as alternate gene silencing products.

For independent verification of GPAM (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-90652A, sc-90652B and sc-90652C.

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

GPAM siRNA (h) is recommended for the inhibition of GPAM 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

GPAM (D-10): sc-398135 is recommended as a control antibody for monitoring of GPAM 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 GPAM gene expression knockdown using RT-PCR Primer: GPAM (h)-PR: sc-90652-PR (20 µl, 473 bp). 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.