SANTA CRUZ BIOTECHNOLOGY, INC.

GFAT1 siRNA (m): sc-60682

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Glutamine:fructose-6-phosphate amidotransferase (GFAT1) is the first and rate-limiting enzyme for the entry of glucose into the hexosamine biosynthesis pathway (HBP) in mammals. GFAT1, a member of the N-terminal nucleophile class of amidotransferases, converts fructose-6-phosphate into N-acetylglucosamine-6-phosphate. Hyperglycemia-induced Insulin resistance, a condition in which exposure to high concentrations of glucose and Insulin results in Insulin resistance, may result from increased glucose metabolism through the HBP. Hypergylcemia-induced Insulin resistance is a characteristic feature of type 2 diabetes. Consequently, GFAT1 is a potential therapeutic target in the treatment of type 2 diabetes.

REFERENCES

1. Nerlich, A.G., et al. 1998. Expression of glutamine:fructose-6-phosphate amidotransferase in human evidence for high variability and distinct regulation in diabetes. Diabetes 47: 170-178.
2. DeHaven, J.E., et al. 2001. A novel variant of mRNA is selectively expressed in striated muscle. Diabetes 50: 2419-2424.
3. Niimi, M., et al. 2001. Identification of GFAT1-L, a novel splice variant of human glutamine: fructose-6-phosphate amidotransferase (GFAT1) that is expressed abundantly in skeletal muscle. J. Hum. Genet. 46: 566-571.
4. Kaneto, H., et al. 2001. Activation of the hexosamine pathway leads to deterioration of pancreatic β-cell function through the induction of oxidative stress. J. Biol. Chem. 276: 31099-31104.
5. Broschat, K.O., et al. 2002. A radiometric assay for glutamine:fructose-6-phosphate amidotransferase. Anal. Biochem. 305: 10-15.
6. Broschat, K.O., et al. 2002. Kinetic characterization of human glutamine-fructose-6-phosphate amidotransferase I: potent feedback inhibition by glucosamine 6-phosphate. J. Biol. Chem. 277: 14764-14770.
7. Chou, K.C. 2004. Molecular therapeutic target for type-2 diabetes. J. Proteome Res. 3: 1284-1288.
8. Elbein, S.C., et al. 2004. Molecular screening of the human glutamine-fructose-6-phosphate amidotransferase 1 (GFPT1) gene and association studies with diabetes and diabetic nephropathy. Mol. Genet. Metab. 82: 321-328.

CHROMOSOMAL LOCATION

Genetic locus: Gfpt1 (mouse) mapping to 6 D1.

PRODUCT

GFAT1 siRNA (m) 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 GFAT1 shRNA Plasmid (m): sc-60682-SH and GFAT1 shRNA (m) Lentiviral Particles: sc-60682-V as alternate gene silencing products.

For independent verification of GFAT1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-60682A, sc-60682B and sc-60682C.

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

GFAT1 siRNA (m) is recommended for the inhibition of GFAT1 expression in mouse 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor GFAT1 gene expression knockdown using RT-PCR Primer: GFAT1 (m)-PR: sc-60682-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Koppe, L., et al. 2016. Urea impairs β cell glycolysis and Insulin secretion in chronic kidney disease. J. Clin. Invest. 126: 3598-3612.

RESEARCH USE

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

PROTOCOLS

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