SANTA CRUZ BIOTECHNOLOGY, INC.

PHKG1 siRNA (h): sc-89501

BACKGROUND

PHKG1 (Phosphorylase kinase subunit γ1), is a subunit of phosphorylase kinase (PHK) that belongs to the Ser/Thr protein kinase family. PHK is a hexadecameric protein composed of four α chains, four β chains, four γ chains and four δ chains. The γ chains are catalytic chains, the α and β chains are regulatory chains and the δ chains are calmodulins. PHKG1 contains two calmodulin-binding domains and one protein kinase domain. As the catalytic chain of PHK, PHKG1 is responsible for catalyzing the phosphorylation and activation of glycogen phosphorylase and therefore it plays an important role in the glycogenolytic pathway. Mutations in the gene encoding PHKG1 can lead to PHK deficiency and result in glycogen storage disease type 9C (GSD9C), also known as autosomal liver glycogenosis.

REFERENCES

1. Hanks, S.K. 1989. Messenger ribonucleic acid encoding an apparent isoform of phosphorylase kinase catalytic subunit is abundant in the adult testis. Mol. Endocrinol. 3: 110-116.
2. Calalb, M.B., et al. 1992. Molecular cloning and enzymatic analysis of the rat homolog of "PhK-γT", an isoform of phosphorylase kinase catalytic subunit. J. Biol. Chem. 267: 1455-1463.
3. Liu, L., et al. 1996. The testis isoform of the phosphorylase kinase catalytic subunit (PhK-γT) plays a critical role in regulation of glycogen mobilization in developing lung. J. Biol. Chem. 271: 11761-11766.
4. Maichele, A.J., et al. 1996. Mutations in the testis/liver isoform of the phosphorylase kinase γ subunit (PHKG2) cause autosomal liver glycogenosis in the gsd rat and in humans. Nat. Genet. 14: 337-340.
5. Burwinkel, B., et al. 1998. Liver glycogenosis due to phosphorylase kinase deficiency: PHKG2 gene structure and mutations associated with cirrhosis. Hum. Mol. Genet. 7: 149-154.
6. Burwinkel, B., et al. 2000. Phosphorylase kinase deficient liver glycogenosis: progression to cirrhosis in infancy associated with PHKG2 mutations (H14.and L225R) J. Med. Genet. 37: 376-377.
7. Burwinkel, B., et al. 2003. Severe phenotype of phosphorylase kinase-deficient liver glycogenosis with mutations in the PHKG2 gene. Pediatr. Res. 54: 834-839.

CHROMOSOMAL LOCATION

Genetic locus: PHKG1 (human) mapping to 7p11.2.

PRODUCT

PHKG1 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 PHKG1 shRNA Plasmid (h): sc-89501-SH and PHKG1 shRNA (h) Lentiviral Particles: sc-89501-V as alternate gene silencing products.

For independent verification of PHKG1 (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-89501A, sc-89501B and sc-89501C.

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

PHKG1 siRNA (h) is recommended for the inhibition of PHKG1 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

PHKG1 (RR-34): sc-100536 is recommended as a control antibody for monitoring of PHKG1 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 PHKG1 gene expression knockdown using RT-PCR Primer: PHKG1 (h)-PR: sc-89501-PR (20 µl, 560 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.

PROTOCOLS

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