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PGAM5 siRNA (m): sc-152184

BACKGROUND

Members of the PGAM (phosphoglycerate mutase) family of proteins are important components of glucose and 2,3-BPGA (2,3-bisphosphoglycerate) metabolism. They are responsible for catalyzing the transfer of phospho groups between the carbon atoms of phosphoglycerates. PGAM5 (phosphoglycerate mutase family member 5), also known as Bcl-x_L-binding protein v68, is a 289 amino acid protein belonging to the BPG-dependent PGAM subfamily. PGAM5 exists as two isoforms produced by alternative splicing events, with isoform two localized to the cytoplasm and isoform one localized to both the cytoplasm and the nucleus. PGAM5 forms a dimer and has been found to interact with Bcl-x_{S/L} and Keap1.

REFERENCES

1. Zhang, J., et al. 2001. Mouse phosphoglycerate mutase M and B isozymes: cDNA cloning, enzyme activity assay and mapping. *Gene* 264: 273-279.
2. Hammond, P.W., et al. 2001. *In vitro* selection and characterization of Bcl-x_L-binding proteins from a mix of tissue-specific mRNA display libraries. *J. Biol. Chem.* 276: 20898-20906.
3. Jin, J., et al. 2004. Proteomic, functional, and domain-based analysis of *in vivo* 14-3-3 binding proteins involved in cytoskeletal regulation and cellular organization. *Curr. Biol.* 14: 1436-1450.
4. de Tauri, P., et al. 2005. Characterization of the first described mutation of human red blood cell phosphoglycerate mutase. *Biochim. Biophys. Acta.* 1740: 403-410.
5. Saavedra, E., et al. 2005. Glycolysis in *Entamoeba histolytica*. Biochemical characterization of recombinant glycolytic enzymes and flux control analysis. *FEBS J.* 272: 1767-1783.
6. Lo, S.C. and Hannink, M. 2006. PGAM5, a Bcl-x_L-interacting protein, is a novel substrate for the redox-regulated Keap1-dependent ubiquitin ligase complex. *J. Biol. Chem.* 281: 37893-37903.

CHROMOSOMAL LOCATION

Genetic locus: Pgam5 (mouse) mapping to 5 F.

PRODUCT

PGAM5 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PGAM5 shRNA Plasmid (m): sc-152184-SH and PGAM5 shRNA (m) Lentiviral Particles: sc-152184-V as alternate gene silencing products.

For independent verification of PGAM5 (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-152184A, sc-152184B and sc-152184C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PGAM5 siRNA (m) is recommended for the inhibition of PGAM5 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 μ M in 66 μ l. 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

PGAM5 (A-3): sc-515880 is recommended as a control antibody for monitoring of PGAM5 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 PGAM5 gene expression knockdown using RT-PCR Primer: PGAM5 (m)-PR: sc-152184-PR (20 μ l). 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.