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PYGL siRNA (m): sc-45922

BACKGROUND

Glycolysis is an evolutionarily conserved series of ten chemical reactions that utilizes eleven enzymes to concomitantly generate pyruvate and ATP from glucose. Phospho-fructose kinase-2/fructose 2,6-bisphosphatase (PFK-2) stimulates the synthesis and degradation of fructose 2,6-bisphosphate. Glycogen phosphorylase (also known as GP) is an allosteric enzyme important in carbohydrate metabolism. Its activity is regulated through either noncovalent binding of metabolites or by covalent modification. Glycogen phosphorylase catalyzes the phosphorylation of glycogen to Glc-1-P. There are three genes which encode the brain, liver and muscle forms of glycogen phosphorylase, PYGB, PYGL and PYGM. Because of its fundamental role in the metabolism of glycogen, glycogen phosphorylase has been a target for the design of inhibitory compounds, which could be valuable in the therapeutic treatment of type 2 diabetes mellitus.

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CHROMOSOMAL LOCATION

Genetic locus: *Pygl* (mouse) mapping to 12 C2.

PRODUCT

PYGL 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 PYGL shRNA Plasmid (m): sc-45922-SH and PYGL shRNA (m) Lentiviral Particles: sc-45922-V as alternate gene silencing products.

For independent verification of PYGL (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-45922A, sc-45922B and sc-45922C.

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

PYGL siRNA (m) is recommended for the inhibition of PYGL 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

PYGL (C3): sc-517597 is recommended as a control antibody for monitoring of PYGL 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 PYGL gene expression knockdown using RT-PCR Primer: PYGL (m)-PR: sc-45922-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.

PROTOCOLS

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