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GK1 siRNA (m): sc-145410

BACKGROUND

As the central structural component of the major classes of biological lipids, triglycerides and phosphatidyl phospholipids, glycerol is an essential intermediate in carbohydrate and lipid metabolism. Glycerol kinases (GKs) function to catalyze the transfer of a phosphate group from ATP to glycerol, thereby forming glycerol phosphate. This intermediate can then be converted to dihydroxyacetone phosphate (DHAP), which is utilized in either glycolysis or gluconeogenesis. Mutations in the genes encoding GK family members can result in glycerol kinase deficiency, which is characterized by hyperglycemia, psychomotor retardation and osteoporosis. GK1 is a 559 amino acid mitochondrial peripheral membrane protein that belongs to the FGGY kinase family and is a key enzyme involved in the regulation of glycerol uptake and metabolism. GK1 shows high expression in kidney, testis and liver and exists as three isoforms, which are produced as a result of alternative splicing events.

REFERENCES

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CHROMOSOMAL LOCATION

Genetic locus: Gyk (mouse) mapping to X C1.

PRODUCT

GK1 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 GK1 shRNA Plasmid (m): sc-145410-SH and GK1 shRNA (m) Lentiviral Particles: sc-145410-V as alternate gene silencing products.

For independent verification of GK1 (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-145410A, sc-145410B and sc-145410C.

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

GK1 siRNA (m) is recommended for the inhibition of GK1 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

GK1 (E-4): sc-398385 is recommended as a control antibody for monitoring of GK1 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 GK1 gene expression knockdown using RT-PCR Primer: GK1 (m)-PR: sc-145410-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.