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SANTA CRUZ BIOTECHNOLOGY, INC.

PFKL siRNA (m): sc-152180

BACKGROUND

Phosphofructokinases (PFKs) are regulatory glycolytic enzymes that catalyze the irreversible conversion of fructose-6-phosphate to fructose-1,6-bisphosphate. Mammalian PFK is a tetramer made up of diverse combinations of three isoenzymes: muscle (PFK-1), liver (PFKL) and platelet (PFKP). PFKL (phosphofructokinase, liver), also referred to as PFK-B (Phosphofructo-1-kinase isozyme B), phosphofructokinase 1 or phosphohexokinase, predominates in organs with active gluconeogenesis, such as liver and kidney. Overexpression of PFKL in transgenic mice results in a diminished glucose-induced Insulin response, which suggests that PFKL may play a role in glucose-induced Insulin secretion. PFKL is expressed at high levels in Down's syndrome (DS) patients, suggesting a possible role for PFKL in the pathogenesis of DS.

REFERENCES

1. Vora, S. and Francke, U. 1981. Assignment of the human gene for liver-type 6-phosphofructokinase isozyme (PFKL) to chromosome 21 by using somatic cell hybrids and monoclonal anti-L antibody. Proc. Natl. Acad. Sci. USA 78(6): 3738-3742. PMID: 6455664
2. Levanon, D., et al. 1986. Genomic clones of the human liver-type phosphofructokinase. Biochem. Biophys. Res. Commun. 141: 374-380.
3. Levanon, D., et al. 1987. Construction of a cDNA clone containing the entire coding region of the human liver-type phosphofructokinase. Biochem. Biophys. Res. Commun. 147: 1182-1187.
4. Elson, A., et al. 1994. Overexpression of liver-type phosphofructokinase (PFKL) in transgenic-PFKL mice: implication for gene dosage in trisomy 21. Biochem. J. 299: 409-415.
5. Knobler, H., et al. 1997. Impaired glucose-induced Insulin response in transgenic mice overexpressing the L-phosphofructokinase gene. Diabetes 46: 1414-1418.
6. Peled-Kamar, M., et al. 1998. Altered brain glucose metabolism in transgenic-PFKL mice with elevated L-phosphofructokinase: *in vivo* NMR studies. Brain Res. 810: 138-145.
7. Wang, Q., et al. 2005. Rapid prenatal detection of Down syndrome by homologous gene quantitative PCR. Zhonghua Yi Xue Yi Chuan Xue Za Zhi 22: 209-211.

CHROMOSOMAL LOCATION

Genetic locus: Pfkl (mouse) mapping to 10 C1.

PRODUCT

PFKL 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 PFKL shRNA Plasmid (m): sc-152180-SH and PFKL shRNA (m) Lentiviral Particles: sc-152180-V as alternate gene silencing products.

For independent verification of PFKL (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-152180A, sc-152180B and sc-152180C.

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

PFKL siRNA (m) is recommended for the inhibition of PFKL 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.

GENE EXPRESSION MONITORING

PFKL (A-6): sc-393713 is recommended as a control antibody for monitoring of PFKL 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 PFKL gene expression knockdown using RT-PCR Primer: PFKL (m)-PR: sc-152180-PR (20 µl). 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.