

HXK I siRNA (h): sc-39044

BACKGROUND

The hexokinases utilize Mg-ATP as a phosphoryl donor to catalyze the first step of intracellular glucose metabolism, the conversion of glucose to glucose-6-phosphate. Four hexokinase isoenzymes have been identified, including hexokinase I (HXK I), hexokinase II (HXK II), hexokinase III (HXK III) and hexokinase IV (HXK IV, also designated glucokinase or GCK). Hexokinases I-III each contain an N-terminal cluster of hydrophobic amino acids. Glucokinase lacks the N-terminal hydrophobic cluster. The hydrophobic cluster is thought to be necessary for membrane binding. This is substantiated by the finding that glucokinase has lower affinity for glucose than do the other hexokinases. HXK I has been shown to be expressed in brain, kidney and heart tissues as well as in hepatoma cell lines. HXK II is involved in the uptake and utilization of glucose by adipose and skeletal tissues. Of the hexokinases, HXK III has the highest affinity for glucose. Glucokinase is expressed in pancreatic β cells where it functions as a glucose sensor, determining the "set point" for Insulin secretion.

REFERENCES

1. Katzen, H.M. and Schimke, R.T. 1965. Multiple forms of hexokinase in the rat: tissue distribution, age dependency, and properties. *Proc. Natl. Acad. Sci. USA* 54: 1218-1225.
2. Arora, K.K., et al. 1990. Glucose phosphorylation in tumor cells. Cloning, sequencing and overexpression in active form of a fulllength cDNA encoding a mitochondrial bindable form of hexokinase. *J. Biol. Chem.* 265: 6481-6488.

CHROMOSOMAL LOCATION

Genetic locus: HK1 (human) mapping to 10q22.1.

PRODUCT

HXK I 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see HXK I shRNA Plasmid (h): sc-39044-SH and HXK I shRNA (h) Lentiviral Particles: sc-39044-V as alternate gene silencing products.

For independent verification of HXK I (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-39044A, sc-39044B and sc-39044C.

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

HXK I siRNA (h) is recommended for the inhibition of HXK I 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 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

HXK I (G-1): sc-46695 is recommended as a control antibody for monitoring of HXK I 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 HXK I gene expression knockdown using RT-PCR Primer: HXK I (h)-PR: sc-39044-PR (20 μ l, 547 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Quach, C.H., et al. 2016. Mild alkalization acutely triggers the warburg effect by enhancing hexokinase activity via voltage-dependent anion channel binding. *PLoS ONE* 11: e0159529.
2. Depaoli, M.R., et al. 2018. Real-time imaging of mitochondrial ATP dynamics reveals the metabolic setting of single cells. *Cell Rep.* 25: 501-512.

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.