

Glut4 siRNA (m): sc-41221

BACKGROUND

Glucose is fundamental to the metabolism of mammalian cells. Its passage across cell membranes is mediated by a family of transporters termed glucose transporters or Gluts. In adipose and muscle tissue, Insulin stimulates a rapid and dramatic increase in glucose uptake, which is largely due to the redistribution of the Insulin-inducible glucose transporter, Glut4. In response to Insulin, Glut4 is quickly shuttled from an intracellular storage site to the plasma membrane, where it binds glucose. In contrast, the ubiquitously expressed glucose transporter Glut1 is constitutively targeted to the plasma membrane and shows a much less dramatic translocation in response to Insulin. Glut1 and Glut4 are twelve-pass transmembrane proteins (12TM) whose carboxy-termini may dictate their cellular localization. Aberrant Glut4 expression has been suggested to contribute to such maladies as obesity and diabetes. Glut4 null mice have shown that while functional Glut4 protein is not required for maintaining normal glucose levels, it is necessary for sustained growth, normal cellular glucose, fat metabolism and prolonged longevity.

REFERENCES

1. Fischbarg, J., et al. 1993. Evidence that facilitative glucose transporters may fold as β -barrels. *Proc. Natl. Acad. Sci. USA* 90: 11658-11662.
2. Livingstone, C., et al. 1995. Hypothalamic Glut4 expression: a glucose- and Insulin-sensing mechanism? *Mol. Endocrinol.* 107: 67-70.
3. Haney, P.M., et al. 1995. Insulin-sensitive targeting of the Glut4 glucose transporter in L6 myoblasts is conferred by its COOH-terminal cytoplasmic tail. *J. Cell Biol.* 129: 641-658.
4. Kandror, K.V., et al. 1995. Expression and compartmentalization of caveolin in adipose cells: coordinate regulation with and structural segregation from Glut4. *J. Cell Biol.* 129: 999-1006.
5. Marsh, B.J., et al. 1995. Molecular regulation of Glut4 targeting in 3T3-L1 adipocytes. *J. Cell Biol.* 130: 1081-1091.
6. Katz, E.B., et al. 1995. Cardiac and adipose tissue abnormalities but not diabetes in mice deficient in Glut4. *Nature* 377: 151-155.
7. Hajdich, E., et al. 1995. Regulation of glucose transporters in cultured rat adipocytes: synergistic effect of insulin and dexamethasone on Glut4 gene expression through promoter activation. *Endocrinology* 136: 4782-4789.

CHROMOSOMAL LOCATION

Genetic locus: Slc2a4 (mouse) mapping to 11 B3.

PRODUCT

Glut4 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 Glut4 shRNA Plasmid (m): sc-41221-SH and Glut4 shRNA (m) Lentiviral Particles: sc-41221-V as alternate gene silencing products.

For independent verification of Glut4 (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-41221A, sc-41221B and sc-41221C.

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

Glut4 siRNA (m) is recommended for the inhibition of Glut4 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

Glut4 (IF8): sc-53566 is recommended as a control antibody for monitoring of Glut4 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 Glut4 gene expression knockdown using RT-PCR Primer: Glut4 (m)-PR: sc-41221-PR (20 μ l, 454 bp). 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.