

PDX-1 siRNA (r): sc-108040

BACKGROUND

Pancreatic duodenal homeobox-1 protein (PDX-1), also designated Insulin promoter factor (IPF1), Insulin upstream factor 1 (IUF1), somatostatin *trans*-activating factor-1 (STF-1) and glucose-sensitive factor (GSF), is a 282 amino acid homeodomain-containing transcription factor present in pancreatic β -cells. PDX-1 is a key regulator of pancreatic islet development and Insulin gene transcription in β -cells. PDX-1 is expressed in all cells at the early stages of development and is mainly restricted to the pancreas and duodenum in adult. HNF-3 β , HNF-1 α and SP1 positively regulate the PDX-1 enhancer element. PDX-1 is also regulated by glucagon-like peptide through activation of adenylyl cyclase, which results in an increase in intracellular cAMP activity. The increased levels of cAMP, and the resulting activation of PKA, lead to an increase in PDX-1 transcription and translocation of the protein to the nuclei of β -cells. PDX-1 binds to the sequence C(C/T) and can heterodimerize with PBX. PDX-1 is phosphorylated by the SAPK2 pathway under high glucose concentrations. Mutations in the PDX-1 gene can cause maturity-onset diabetes of the young and pancreatic agenesis. The gene which encodes PDX-1 maps to human chromosome 13q12.1.

CHROMOSOMAL LOCATION

Genetic locus: Pdx1 (rat) mapping to 12p11.

PRODUCT

PDX-1 siRNA (r) 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 PDX-1 shRNA Plasmid (r): sc-108040-SH and PDX-1 shRNA (r) Lentiviral Particles: sc-108040-V as alternate gene silencing products.

For independent verification of PDX-1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-108040A, sc-108040B and sc-108040C.

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

PDX-1 siRNA (r) is recommended for the inhibition of PDX-1 expression in rat cells.

PROTOCOLS

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

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

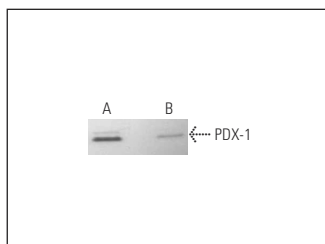
PDX-1 (B-11): sc-390792 is recommended as a control antibody for monitoring of PDX-1 gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

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 PDX-1 gene expression knockdown using RT-PCR Primer: PDX-1 (r)-PR: sc-108040-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



PDX-1 siRNA (r): sc-108040. Immunoblotting of PDX-1 expression using PDX-1 antibody on INS1 cell extracts treated with Control siRNA-A: sc-37007 (A) and PDX-1 siRNA: sc-108040 (B).

SELECT PRODUCT CITATIONS

- Kim, J.Y., et al. 2014. Chronic ethanol consumption inhibits glucokinase transcriptional activity by Atf3 and triggers metabolic syndrome *in vivo*. J. Biol. Chem. 289: 27065-27079.

RESEARCH USE

For research use only, not for use in diagnostic procedures.