SANTA CRUZ BIOTECHNOLOGY, INC.

IRS-1 siRNA (h): sc-29376

BACKGROUND

The Insulin receptor substrate-1 (IRS-1), a protein major substrate of the Insulin receptor, is phosphorylated in response to stimulation of cells by Insulin, Insulin-like growth factor 1 (IGF-1) and interleukin 4 (IL-4). IRS-1 is phosphorylated on serine, threonine and tyrosine residues in a variety of tissues. An Insulin-sensitive serine/threonine kinase casein kinase II mediates a portion of the Insulin-stimulated serine/threonine phosphorylation of overexpressed IRS-1 *in vivo*. Thr 502 is identified as the major casein kinase II-catalyzed phosphorylation site in rat IRS-1, and Ser 99 is an additional phosphorylation site catalyzed by casein kinase II. Thus, casein kinase II-catalyzed phosphorylation of IRS-1 may be a component of the intracellular Insulin signaling cascade. IRS-1 contains three putative binding sites for 14-3-3 (Ser 270, Ser 374 and Ser 641) and the motif around Ser 270 is located in the phosphotyrosine binding domain of IRS-1, which is responsible for the interaction with the Insulin receptor. The association of 14-3-3 with IRS-1 increases significantly upon treatment with okadaic acid, a potent serine/threonine phosphatase inhibitor. Therefore, the association of 14-3-3 protein may play a role in the regulation of Insulin sensitivity by interrupting the association between the Insulin receptor and IRS-1.

CHROMOSOMAL LOCATION

Genetic locus: IRS1 (human) mapping to 2q36.3.

PRODUCT

IRS-1 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see IRS-1 shRNA Plasmid (h): sc-29376-SH and IRS-1 shRNA (h) Lentiviral Particles: sc-29376-V as alternate gene silencing products.

For independent verification of IRS-1 (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-29376A, sc-29376B and sc-29376C.

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

IRS-1 siRNA (h) is recommended for the inhibition of IRS-1 expression in human 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 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.

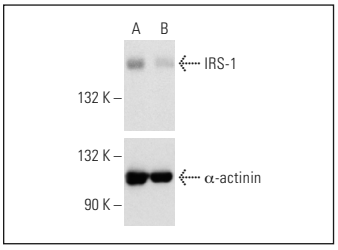
GENE EXPRESSION MONITORING

IRS-1 (H-7): sc-515017 is recommended as a control antibody for monitoring of IRS-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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor IRS-1 gene expression knockdown using RT-PCR Primer: IRS-1 (h)-PR: sc-29376-PR (20 µl, 411 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



IRS-1 siRNA (h): sc-29376. Western blot analysis of IRS-1 expression in non-transfected control (**A**) and IRS-1 siRNA transfected (**B**) HeLa cells. Blot probed with IRS-1 (C-20): sc-559. α-actinin (H-2): sc-17829 used as specificity and loading control.

SELECT PRODUCT CITATIONS

1. Asano, T., et al. 2005. Insulin receptor substrate is a mediator of phosphoinositide 3-kinase activation in quiescent pancreatic cancer cells. Cancer Res. 65: 9164-9168.
2. Tan, X., et al. 2016. Phosphoproteome analysis of invasion and metastasis-related factors in pancreatic cancer cells. PLoS ONE 11: e0152280.
3. Sun, C., et al. 2024. Mitigation of gestational diabetes-induced endothelial dysfunction through FGF21-NRF2 pathway activation involving L-Cystine. Biochim Biophys Acta Mol Basis Dis. 1870: 167329.

RESEARCH USE

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