SANTA CRUZ BIOTECHNOLOGY, INC.

BRIDGE-1 siRNA (h): sc-105127

BACKGROUND

BRIDGE-1, a protein homologous to a previously cloned proteasome subunit p27 is important in regulating Insulin and other islet genes in the pancreas. BRIDGE-1 is highly expressed in pancreatic b-cells and is predominantly located in the nucleus, although lower levels are expressed in the cytoplasm. BRIDGE-1 contains a conserved PDZ-like domain that mediates protein-protein interactions in a variety of intracellular signaling processes, including the transactivational activity of E2A. One mechanism of the activation of gene transcription in pancreatic B-cells is the interaction of E2A with coactivating proteins such as CBP, p300 and BRIDGE-1. The interaction of E12 and E47, members of the E2A family of transcription factors, with the PDZ-domain of BRIDGE-1 suggest a novel mechanism for Insulin gene regulation.

REFERENCES

1. German, M.S., et al. 1994. The Insulin gene contains multiple transcriptional elements that respond to glucose. Mol. Cell. Biol. 14: 4067-4075.
2. Cordier-Bussat, M., et al. 1995. Homologous DNA sequences and cellular factors are implicated in the control of Glucagon and Insulin gene expression. Mol. Cell. Biol. 15: 3904-3916.
3. Saras, J., et al. 1996. PDZ domains bind carboxy-terminal sequences of target proteins. Trends Biochem. Sci. 21: 455-458.
4. Eckner, R., et al. 1996. Interaction and functional collaboration of p300/CBP and bHLH proteins in muscle and B-cell differentiation. Genes Dev. 10: 2478-2490.
5. Watanabe, T.K., et al. 1998. cDNA cloning and characterization of a human proteasomal modulator subunit, p27 (PSMD9). Genomics 50: 241-250.
6. Qiu, Y., et al. 1998. p300 mediates transcriptional stimulation by the basic helix-loop-helix activators of the Insulin gene. Mol. Cell. Biol. 18: 2957-2964.
7. Thomas, M.K., et al. 1999. BRIDGE-1, a novel PDZ-domain coactivator of E2A-mediated regulation of Insulin gene transcription. Mol. Cell. Biol. 19: 8492-8504.

CHROMOSOMAL LOCATION

Genetic locus: PSMD9 (human) mapping to 12q24.31.

PRODUCT

BRIDGE-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 BRIDGE-1 shRNA Plasmid (h): sc-105127-SH and BRIDGE-1 shRNA (h) Lentiviral Particles: sc-105127-V as alternate gene silencing products.

For independent verification of BRIDGE-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-105127A, sc-105127B and sc-105127C.

PROTOCOLS

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

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

BRIDGE-1 siRNA (h) is recommended for the inhibition of BRIDGE-1 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 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

BRIDGE-1 (D-4): sc-376363 is recommended as a control antibody for monitoring of BRIDGE-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 BRIDGE-1 gene expression knockdown using RT-PCR Primer: BRIDGE-1 (h)-PR: sc-105127-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.