

PACAP siRNA (h): sc-39530

BACKGROUND

Glucagon is a pancreatic hormone that functions as an antagonist to Insulin, stimulating the conversion of glycogen to glucose and increasing blood sugar levels. Glucagon-like peptide-1 (GLP-1), Glucagon-like peptide-2 (GLP-2), VIP (vasoactive intestinal peptide) and PACAP (pituitary adenylate cyclase activating polypeptide) are members of the glucagon family of hormones. GLP-1 functions as a transmitter in the central nervous system, inhibiting feeding and drinking behavior, whereas GLP-2 is a stimulator of intestinal epithelial growth. VIP causes vasodilation resulting in the lowering of blood pressure. PACAP is abundant in the hypothalamus and has been shown to increase the synthesis of several hormones, including growth hormone.

REFERENCES

1. Rouille, Y., et al. 1995. Differential processing of proglucagon by the subtilisin-like prohormone convertases PC2 and PC3 to generate either glucagon or glucagon-like peptide. *J. Biol. Chem.* 270: 26488-26496.
2. Moens, K., et al. 1996. Expression and functional activity of glucagon, glucagon-like peptide I, and glucose-dependent Insulinotropic peptide receptors in rat pancreatic islet cells. *Diabetes* 45: 257-261.
3. Scrocchi, L.A., et al. 1996. Glucose intolerance but normal satiety in mice with a null mutation in the glucagon-like peptide 1 receptor gene. *Nat. Med.* 2: 1254-1258.
4. Jiang, S., et al. 1997. Vasoactive intestinal peptide (VIP) stimulates *in vitro* growth of VIP-1 receptor-bearing human pancreatic adenocarcinoma-derived cells. *Cancer Res.* 57: 1475-1480.
5. Bollen, M., et al. 1998. Specific features of glycogen metabolism in the liver. *Biochem. J.* 336: 19-31.
6. Martinez-Fuentes, A.J., et al. 1998. Pituitary adenylate cyclase-activating polypeptide (PACAP) 38 and PACAP27 activate common and distinct intracellular signaling pathways to stimulate growth hormone secretion from porcine somatotropes. *Endocrinology* 139: 5116-5124.

CHROMOSOMAL LOCATION

Genetic locus: ADCYAP1 (human) mapping to 18p11.32.

PRODUCT

PACAP 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 PACAP shRNA Plasmid (h): sc-39530-SH and PACAP shRNA (h) Lentiviral Particles: sc-39530-V as alternate gene silencing products.

For independent verification of PACAP (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-39530A, sc-39530B and sc-39530C.

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 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

PACAP siRNA (h) is recommended for the inhibition of PACAP 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

PACAP (F-2): sc-166180 is recommended as a control antibody for monitoring of PACAP 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 PACAP gene expression knockdown using RT-PCR Primer: PACAP (h)-PR: sc-39530-PR (20 μ l). 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.