

PCSK1 siRNA (r): sc-270276

BACKGROUND

PCSK1 (proprotein convertase subtilisin/kexin type 1), also known as PC1 (prohormone convertase 1), PC3, NEC1 (neuroendocrine convertase 1) or SPC3, is a member of the subtilisin-like proprotein convertase family that plays an important role in the processing of hormones such as Renin, Dynorphin, POMC, Synenkephalin, Insulin and Somatostatin. PCSK1 localizes to the cytoplasm and, using calcium as a cofactor, specifically catalyzes a hydrolysis reaction that releases protein hormones, Renin and neuropeptides from their corresponding precursors. PCSK1 is involved in the regulation of Insulin biosynthesis and functions as a type I proinsulin-processing enzyme. Mutations in the gene encoding PCSK1 can lead to PC1 deficiency, a disorder characterized by hypogonadism, obesity, reactive hypoglycemia, hypoadrenalism and small-intestinal absorptive dysfunction. Various isoforms exist for PCSK1 due to alternative splicing events.

REFERENCES

- Seidah, N.G., et al. 1991. Chromosomal assignments of the genes for neuroendocrine convertase PC1 (NEC1) to human 5q15-21, neuroendocrine convertase PC2 (NEC2) to human 20p11.1-11.2, and furin (mouse 7[D1-E2] region). *Genomics* 11: 103-107.
- Seidah, N.G., et al. 1992. The cDNA sequence of the human pro-hormone and pro-protein convertase PC1. *DNA Cell Biol.* 11: 283-289.
- Ohagi, S., et al. 1996. Human prohormone convertase 3 gene: exon-intron organization and molecular scanning for mutations in Japanese subjects with NIDDM. *Diabetes* 45: 897-901.
- Boudreault, A., et al. 1998. Proprotein convertase PC1/3-related peptides are potent slow tight-binding inhibitors of murine PC1/3 and Hfurin. *J. Biol. Chem.* 273: 31574-31580.
- Qian, Y., et al. 2000. The C-terminal region of proSAAS is a potent inhibitor of prohormone convertase 1. *J. Biol. Chem.* 275: 23596-23601.
- Arnautova, I., et al. 2003. The prohormone processing enzyme PC3 is a lipid raft-associated transmembrane protein. *Biochemistry* 42: 10445-10455.
- Jackson, R.S., et al. 2003. Small-intestinal dysfunction accompanies the complex endocrinopathy of human proprotein convertase 1 deficiency. *J. Clin. Invest.* 112: 1550-1560.

CHROMOSOMAL LOCATION

Genetic locus: *Pcsk1* (rat) mapping to 2q11-q12.

PRODUCT

PCSK1 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 PCSK1 shRNA Plasmid (r): sc-270276-SH and PCSK1 shRNA (r) Lentiviral Particles: sc-270276-V as alternate gene silencing products.

For independent verification of PCSK1 (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-270276A, sc-270276B and sc-270276C.

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

PCSK1 siRNA (r) is recommended for the inhibition of PCSK1 expression in rat 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

PCSK1 (J-18): sc-100578 is recommended as a control antibody for monitoring of PCSK1 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 PCSK1 gene expression knockdown using RT-PCR Primer: PCSK1 (r)-PR: sc-270276-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.

PROTOCOLS

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