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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

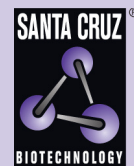
T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



Pax-4 shRNA (h) Lentiviral Particles: sc-43998-V

BACKGROUND

Pax-4 (paired box gene 4) protein influences normal differentiation of insulin-producing β cells and influences normal pancreatic islet development. Pax-4 protein is a transcriptional repressor that binds to a common *cis* element in the glucagon, insulin and somatostatin promoters. Mouse Pax-4 transcript is present in pancreatic islets, and the islet β cell lines MIN6, β TC, and NIT1. Differentiation of endoderm-derived endocrine pancreas is mediated through Pax-4 and Pax-6. Pax-4 may act as a Pax-6 repressor due to the competition for binding sites and lower transactivation potential of Pax-4. The human Pax-4 gene encodes a deduced 350 amino acid protein that is 80% identical to the deduced mouse Pax-4 protein.

REFERENCES

1. Matsushita, T., et al. 1998. Molecular cloning of mouse paired box containing gene Pax-4 from an islet β cell line and deduced sequence of human Pax-4. *Biochem. Biophys. Res. Commun.* 242: 176-180.
2. Larsson, L.I., et al. 1998. Pax-4 and -6 regulate gastrointestinal endocrine cell development. *Mech. Dev.* 79: 153-159.
3. Kalousová, A., et al. 1999. DNA binding and transactivating properties of the paired and homeobox protein Pax-4. *Biochem. Biophys. Res. Commun.* 259: 510-518.
4. Ritz-Laser, B., et al. 2002. The pancreatic β cell-specific transcription factor Pax-4 inhibits Glucagon gene expression through Pax-6. *Diabetologia* 45: 97-107.
5. Kemp, D.M., et al. 2003. Regulation of Pax-4 paired homeodomain gene by neuron-restrictive silencer factor. *J. Biol. Chem.* 278: 35057-35062.

CHROMOSOMAL LOCATION

Genetic locus: PAX4 (human) mapping to 7q32.1.

PRODUCT

Pax-4 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Pax-4 siRNA (h): sc-43998 and Pax-4 shRNA Plasmid (h): sc-43998-SH as alternate gene silencing products.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

APPLICATIONS

Pax-4 shRNA (h) Lentiviral Particles is recommended for the inhibition of Pax-4 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Pax-4 (S-15): sc-27832 is recommended as a control antibody for monitoring of Pax-4 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 (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Pax-4 gene expression knockdown using RT-PCR Primer: Pax-4 (h)-PR: sc-43998-PR (20 μ l, 497 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

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