



Pax-3 siRNA (h): sc-38747

BACKGROUND

Pax genes contain paired domains that share strong homology to genes in *Drosophila* which are involved in programming early development. The product of the PAX3 gene is a DNA-binding protein expressed during early neurogenesis. Pax-3 is a protein containing both a paired domain and a paired-type homeodomain. During early neurogenesis, Pax-3 expression is limited to mitotic cells in the ventricular zone of the developing spinal cord and to distinct regions in the hindbrain, midbrain and diencephalon. In 10-12 day embryos, expression of Pax-3 is also seen in neural crest cells of the developing spinal ganglia, the craniofacial mesectoderm and in limb mesenchyme. Mutations in the MITF and PAX3 genes, encoding transcription factors, are responsible for Waardenburg syndrome II (WSII) and WSI/WSIII, respectively.

REFERENCES

1. Goulding, M.D., et al. 1991. Pax-3, a novel murine DNA binding protein expressed during early neurogenesis. *EMBO J.* 10: 1135-1147.
2. Tassabehji, M., et al. 1992. Waardenburg's syndrome patients have mutations in the human homologue of the paired box gene. *Nature* 355: 635-636.
3. Hoth, C.F., et al. 1993. Mutations in the paired domain of the human PAX3 gene cause Klein-Waardenburg syndrome (WS-III) as well as Waardenburg syndrome type I (WS-I). *Am. J. Hum. Genet.* 52: 455-462.
4. Stapleton, P., et al. 1993. Chromosomal localization of seven PAX genes and cloning of a novel family member, PAX9. *Nat. Genet.* 3: 292-298.
5. Tsukamoto, K., et al. 1994. Isolation of two isoforms of the PAX3 gene transcripts and their tissue-specific alternative expression in human adult tissues. *Hum. Genet.* 93: 270-274.
6. Watanabe, A., et al. 1998. Epistatic relationship between Waardenburg syndrome genes MITF and PAX3. *Nat. Genet.* 18: 283-286.
7. Wachtel, M., et al. 2004. Gene expression signatures identify rhabdomyosarcoma subtypes and detect a novel t(2;2)(q35;p23) translocation fusing PAX3 to NCOA1. *Cancer Res.* 64: 5539-5545.
8. SWISS-PROT/TrEMBL (P23760). World Wide Web URL: <http://www.expasy.ch/sprot/sprot-top.html>

CHROMOSOMAL LOCATION

Genetic locus: PAX3 (human) mapping to 2q36.1.

PRODUCT

Pax-3 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 Pax-3 shRNA Plasmid (h): sc-38747-SH and Pax-3 shRNA (h) Lentiviral Particles: sc-38747-V as alternate gene silencing products.

For independent verification of Pax-3 (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-38747A, sc-38747B and sc-38747C.

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

Pax-3 siRNA (h) is recommended for the inhibition of Pax-3 expression in human cells.

Pax-3 (L-14): sc-34918 is recommended as a control antibody for Western Blotting (starting dilution 1:100, dilution range 1:100-1:1,000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) protein detection using the recommended secondary reagents listed below.

RECOMMENDED SECONDARY REAGENTS

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.

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 60 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. Semi-quantitative RT-PCR may be performed using RT-PCR Primer: Pax-3 (h)-PR: sc-38747-PR (20 μ l, 576 bp). 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 or our catalog for detailed protocols and support products.