SANTA CRUZ BIOTECHNOLOGY, INC.

Pax-1 siRNA (m): sc-38744

BACKGROUND

Pax genes contain paired domains with strong homology to genes in *Drosophila* that are involved in programming early development. Pax-1 is a sequence-specific DNA binding-protein with transcriptional activating properties. The expression pattern of Pax-1 during mouse embryogenesis indicates that it may play an important role in the development of the vertebral column. The autosomal recessive mutation "undulated" (un) in the mouse exhibits vertebral anomalies along the entire rostrocaudal axis and is associated with a point mutation (G-to-A transition) at position 15 leading to a gly-to-ser replacement in a highly conserved region of the paired box of Pax-1. Pax-1 is required for the normal development of these three skeletal elements: the vertebral column, sternum, and scapula. Mice who are doubly heterozygous for the mutants "undulated" and "Patch" have a phenotype reminiscent of an extreme form of spina bifida occulta in humans. The gene which encodes Pax-1 maps to human chromosome 20p11.22.

REFERENCES

1. Deutsch, U., et al. 1988. Pax-1, a member of a paired box homologous murine gene family, is expressed in segmented structures during development. Cell 53: 617-625.
2. Lichter, P., et al. 1990. High-resolution mapping of human chromosome 11 by *in situ* hybridization with cosmid clones. Science 247: 64-69.
3. Chalepakis, G., et al. 1991. The molecular basis of the undulated/Pax-1 mutation. Cell 66: 873-884.
4. Helwig, U., et al. 1995. Interaction between undulated and Patch leads to an extreme form of spina bifida in double-mutant mice. Nat. Genet. 11: 60-63.
5. David, K.M., et al. 1999. The dysmorphic cervical spine in Klippel-Feil syndrome: interpretations from developmental biology. Neurosurg. Focus 6: E1.
6. Tang, L.S., et al. 2003. Neural and orofacial defects in Folp1 knockout mice [corrected]. Birth Defects Res. A, Clin. Mol. Teratol. 67: 209-218.
7. Wilson-Rawls, J., et al. 2004. Paraxis is a basic helix-loop-helix protein that positively regulates transcription through binding to specific E-box elements. J. Biol. Chem. 279: 37685-37692.

CHROMOSOMAL LOCATION

Genetic locus: Pax1 (mouse) mapping to 2 G2.

PRODUCT

Pax-1 siRNA (m) 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 Pax-1 shRNA Plasmid (m): sc-38744-SH and Pax-1 shRNA (m) Lentiviral Particles: sc-38744-V as alternate gene silencing products.

For independent verification of Pax-1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-38744A, sc-38744B and sc-38744C.

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

Pax-1 siRNA (m) is recommended for the inhibition of Pax-1 expression in mouse 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

Pax (D-7): sc-514352 is recommended as a control antibody for monitoring of Pax-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).

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Pax-1 gene expression knockdown using RT-PCR Primer: Pax-1 (m)-PR: sc-38744-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.

PROTOCOLS

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