



# Pax-6 shRNA (m) Lentiviral Particles: sc-36196-V

## BACKGROUND

Pax genes contain paired domains with strong homology to genes in *Drosophila* which are involved in programming early development. Lesions in the Pax-6 gene accounts for most cases of aniridia, a congenital malformation of the eye, chiefly characterized by iris hypoplasia, which can cause blindness. Pax-6 is involved in other anterior segment malformations besides aniridia, such as Peters' anomaly, a major error in the embryonic development of the eye with corneal clouding with variable iridolenticulocorneal adhesions. The Pax-6 gene encodes a transcriptional regulator that recognizes target genes through its paired-type DNA-binding domain. The paired domain is composed of two distinct DNA-binding subdomains, the amino-terminal subdomain and the carboxy-terminal subdomain, which bind respective consensus DNA sequences. The human Pax-6 gene produces two alternatively spliced isoforms that have the distinct structure of the paired domain.

## REFERENCES

- Hanson, I.M., et al. 1993. Pax-6 mutations in aniridia. *Hum. Mol. Genet.* 2: 915-920.
- Hanson, I.M., et al. 1994. Mutations at the Pax-6 locus are found in heterogeneous anterior segment malformations including Peters' anomaly. *Nat. Genet.* 6: 168-173.
- Azuma, N., et al. 1999. Missense mutation in the alternative splice region of the Pax-6 gene in eye anomalies. *Am. J. Hum. Genet.* 65: 656-663.
- Fic, W., et al. 2007. Eye development under the control of SRp55/B52-mediated alternative splicing of *eyeless*. *PLoS ONE* 2: e253.
- Yan, Q., et al. 2007. Protein phosphatase-1 modulates the function of Pax-6, a transcription factor controlling brain and eye development. *J. Biol. Chem.* 282: 13954-13965.

## CHROMOSOMAL LOCATION

Genetic locus: Pax6 (mouse) mapping to 2 E3.

## PRODUCT

Pax-6 shRNA (m) 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  $\mu$ l frozen stock containing  $1.0 \times 10^6$  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-6 siRNA (m): sc-36196 and Pax-6 shRNA Plasmid (m): sc-36196-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-6 shRNA (m) Lentiviral Particles is recommended for the inhibition of Pax-6 expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200  $\mu$ l frozen viral stock containing  $1.0 \times 10^6$  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-6 (H-295): sc-11357 is recommended as a control antibody for monitoring of Pax-6 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 goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (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 goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (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-6 gene expression knockdown using RT-PCR Primer: Pax-6 (m)-PR: sc-36196-PR (20  $\mu$ l, 494 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](http://www.scbt.com) or our catalog for detailed protocols and support products.