

## ALX3 siRNA (h): sc-38645

### BACKGROUND

Aristaless-related genes are a group of paired-related homeobox genes which play a role in regulating vertebrate embryogenesis. The homeodomain transcription factor aristaless-like 3 (ALX3) is expressed in mouse embryos from eight days of gestation, predominantly in neural crest-derived mesenchyme and in lateral plate mesoderm. Expression analysis of human and mouse tissue reveals predominant ALX3 expression in brain tissue. The Alx3 gene maps to chromosome 1p13.3 and encodes a 343 amino acid protein. Preferential methylation of Alx3 occurs in advanced-stage neuroblastoma and may repress ALX3 expression. Treatment with the methylation inhibitor 5-aza-2'-deoxycytidine restores ALX3 expression. Alx3<sup>-/-</sup> mice lack a phenotype distinct from wild-type mice, however Alx3/Alx4 double mutants demonstrate severe craniofacial abnormalities not present in Alx4 single mutants. Specifically, Alx3/Alx4 double mutant newborn mice have cleft nasal regions in addition to malformation of other neural crest-derived skull structures.

### REFERENCES

1. Ten Berge, D., et al. 1998. Mouse Alx3: an aristaless-like homeobox gene expressed during embryogenesis in ectomesenchyme and lateral plate mesoderm. *Dev. Biol.* 199: 11-25.
2. Meijlink, F., et al. 1999. Vertebrate aristaless-related genes. *Int. J. Dev. Biol.* 43: 651-663.
3. Beverdam, A., et al. 2001. Severe nasal clefting and abnormal embryonic apoptosis in Alx3/Alx4 double mutant mice. *Development* 128: 3975-3986.
4. Online Mendelian Inheritance in Man, OMIM™. 2001. Johns Hopkins University, Baltimore, MD. MIM Number: 606014. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
5. Wimmer, K., et al. 2002. Combined restriction landmark genomic scanning and virtual genome scans identify a novel human homeobox gene, ALX3, that is hypermethylated in neuroblastoma. *Genes Chromosomes Cancer* 33: 285-294.

### CHROMOSOMAL LOCATION

Genetic locus: ALX3 (human) mapping to 1p13.3.

### PRODUCT

ALX3 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ALX3 shRNA Plasmid (h): sc-38645-SH and ALX3 shRNA (h) Lentiviral Particles: sc-38645-V as alternate gene silencing products.

For independent verification of ALX3 (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-38645A, sc-38645B and sc-38645C.

### PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.

### 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

### APPLICATIONS

ALX3 siRNA (h) is recommended for the inhibition of ALX3 expression in human 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  $\mu$ M in 66  $\mu$ 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.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ALX3 gene expression knockdown using RT-PCR Primer: ALX3 (h)-PR: sc-38645-PR (20  $\mu$ 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.