

## IRX3 siRNA (h): sc-106756

### BACKGROUND

The Iroquois homeobox gene family of transcription factors regulate aspects of embryonic development including anterior/posterior and dorsal/ventral axis patterning in the central nervous system. The Iroquois family are clustered on two loci, IRXA and IRXB, which map to chromosomes 8 and 13 in mice. The IRXA group includes IRX, IRX2 and IRX4; the IRXB group comprises IRX3, IRX5 and IRX6. IRX1 and IRX2 are both widely expressed during development in the lung epithelium and also in the ventricular septum. IRX1 and IRX2 also play a role in digit formation (E11.5-E14.5). The IRX gene family members are each expressed in a distinct pattern during mouse heart development. Specifically, IRX1 and IRX2 are expressed in the ventricular septum and IRX3 is expressed in the ventricular trabeculated myocardium. In addition, IRX4 is expressed in the linear heart tube and the AV canal; IRX5 is expressed in the endocardium lining the ventricular and atrial myocardium. Furthermore, the IRX4 gene may modulate cardiac development and function. Although the heart of IRX4<sup>-</sup> mice appears to develop normally, adult IRX4<sup>-</sup> mice exhibit cardiomyopathy, including cardiac hypertrophy and decreased contractility.

### REFERENCES

1. Christoffels, V.M., et al. 2000. Patterning the embryonic heart: identification of five mouse Iroquois homeobox genes in the developing heart. *Dev. Biol.* 224: 263-274.
2. Mummenhoff, J., et al. 2001. Expression of IRX6 during mouse morphogenesis. *Mech. Dev.* 103: 193-195.
3. Becker, M.B., et al. 2001. IRX1 and IRX2 expression in early lung development. *Mech. Dev.* 106: 155-158.
4. Zulch, A., et al. 2001. Expression pattern of IRX1 and IRX2 during mouse digit development. *Mech. Dev.* 106: 159-162.

### CHROMOSOMAL LOCATION

Genetic locus: IRX3 (human) mapping to 16q12.2.

### PRODUCT

IRX3 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 IRX3 shRNA Plasmid (h): sc-106756-SH and IRX3 shRNA (h) Lentiviral Particles: sc-106756-V as alternate gene silencing products.

For independent verification of IRX3 (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-106756A, sc-106756B and sc-106756C.

### RESEARCH USE

For research use only, not for use in diagnostic procedures.

### 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

IRX3 siRNA (h) is recommended for the inhibition of IRX3 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.

### GENE EXPRESSION MONITORING

IRX3 (G-6): sc-166877 is recommended as a control antibody for monitoring of IRX3 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  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-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  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 IRX3 gene expression knockdown using RT-PCR Primer: IRX3 (h)-PR: sc-106756-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.