

## Xin $\alpha$ siRNA (m): sc-63227

### BACKGROUND

Xin $\alpha$ , also known as CMYA1 (cardiomyopathy-associated protein 1) or XIRP1 (Xin actin-binding repeat containing 1), is a 1,843 amino acid protein that functions to protect Actin filaments from depolymerization. Colocalized to the cell junction with actin stress fibers, Xin $\alpha$  contains 15 Xin repeats and interacts with several proteins including  $\beta$ -catenin, Filamin and VASP (vasodilator-stimulated phosphoprotein). The Xin repeats with the protein are thought to stabilize actin-based cytoskeletons and may help to crosslink microfilaments with actin networks. Xin $\alpha$  shares 78% similarity with its mouse counterpart and is expressed in the heart as three alternatively spliced isoforms designated A, B and C. In mice, Xin $\alpha$  is essential for proper heart tube formation and correct cardiac looping, suggesting that the human homolog may have similar functions.

### REFERENCES

1. Wang, D.Z., et al. 1999. Requirement of a novel gene, Xin, in cardiac morphogenesis. *Development* 126: 1281-1294.
2. Sinn, H.W., et al. 2002. Localization of the novel Xin protein to the adherens junction complex in cardiac and skeletal muscle during development. *Dev. Dyn.* 225: 1-13.
3. Online Mendelian Inheritance in Man, OMIM<sup>™</sup>. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 609777. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Pacholsky, D., et al. 2004. Xin repeats define a novel actin-binding motif. *J. Cell Sci.* 117: 5257-5268.
5. Jung-Ching Lin, J., et al. 2005. Structure, expression, and function of a novel intercalated disc protein, Xin. *J. Med. Sci.* 25: 215-222.
6. van der Ven, P.F., et al. 2006. Unusual splicing events result in distinct Xin isoforms that associate differentially with filamin c and Mena/VASP. *Exp. Cell Res.* 312: 2154-2167.

### CHROMOSOMAL LOCATION

Genetic locus: Xirp1 (mouse) mapping to 9 F4.

### PRODUCT

Xin $\alpha$  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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Xin $\alpha$  shRNA Plasmid (m): sc-63227-SH and Xin $\alpha$  shRNA (m) Lentiviral Particles: sc-63227-V as alternate gene silencing products.

For independent verification of Xin $\alpha$  (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-63227A, sc-63227B and sc-63227C.

### 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

Xin $\alpha$  siRNA (m) is recommended for the inhibition of Xin $\alpha$  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  $\mu$ 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

Xin $\alpha$  (D-8): sc-166658 is recommended as a control antibody for monitoring of Xin $\alpha$  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<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> 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<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

### RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Xin $\alpha$  gene expression knockdown using RT-PCR Primer: Xin $\alpha$  (m)-PR: sc-63227-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.