

# Fibulin-4 siRNA (h): sc-75017

## BACKGROUND

Fibulin proteins contribute to normal development of elastic fiber systems in various types of organs that require elasticity, such as vasculature, lung and skin. Fibulin-4, also known as EFEMP2 (EGF-containing fibulin-like extracellular matrix protein 2), MBP1 or UPH1 is a 443 amino acid secreted protein that contains six EGF-like calcium-binding domains and belongs to the fibulin family. Expressed ubiquitously with highest expression in heart, Fibulin-4 is essential for connective tissue development and elastic fiber formation, and may also play an important role in vascular patterning and collagen biosynthesis. Defects in the gene encoding Fibulin-4 are associated with autosomal recessive cutis laxa type I (CL type I), a connective tissue disorder that is inherited in both an autosomal dominant and an autosomal recessive manner and is characterized by inelastic tissue in all affected areas of the body.

## REFERENCES

1. Giltay, R., et al. 1999. Sequence, recombinant expression and tissue localization of two novel extracellular matrix proteins, Fibulin-3 and Fibulin-4. *Matrix Biol.* 18: 469-480.
2. Katsanis, N., et al. 2000. Isolation of a paralog of the Doyme honeycomb retinal dystrophy gene from the multiple retinopathy critical region on 11q13. *Hum. Genet.* 106: 66-72.
3. Gallagher, W.M., et al. 2001. Human Fibulin-4: analysis of its biosynthetic processing and mRNA expression in normal and tumour tissues. *FEBS Lett.* 489: 59-66.
4. Toto, L., et al. 2002. Genetic heterogeneity in Malattia Leventinese. *Clin. Genet.* 62: 399-403.
5. Huchtagowder, V., et al. 2006. Fibulin-4: a novel gene for an autosomal recessive cutis laxa syndrome. *Am. J. Hum. Genet.* 78: 1075-1080.
6. Xiang, Y., et al. 2006. Fibulin-4 is a target of autoimmunity predominantly in patients with osteoarthritis. *J. Immunol.* 176: 3196-3204.
7. McLaughlin, P.J., et al. 2006. Targeted disruption of Fibulin-4 abolishes elastogenesis and causes perinatal lethality in mice. *Mol. Cell. Biol.* 26: 1700-1709.

## CHROMOSOMAL LOCATION

Genetic locus: EFEMP2 (human) mapping to 11q13.1.

## PRODUCT

Fibulin-4 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 Fibulin-4 shRNA Plasmid (h): sc-75017-SH and Fibulin-4 shRNA (h) Lentiviral Particles: sc-75017-V as alternate gene silencing products.

For independent verification of Fibulin-4 (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-75017A, sc-75017B and sc-75017C.

## 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

Fibulin-4 siRNA (h) is recommended for the inhibition of Fibulin-4 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

Fibulin-4 (2C8): sc-293492 is recommended as a control antibody for monitoring of Fibulin-4 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 Fibulin-4 gene expression knockdown using RT-PCR Primer: Fibulin-4 (h)-PR: sc-75017-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## SELECT PRODUCT CITATIONS

1. Choudhury, R., et al. 2009. Differential regulation of elastic fiber formation by Fibulin-4 and -5. *J. Biol. Chem.* 284: 24553-24567.

## RESEARCH USE

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