

# FREM2 siRNA (m): sc-75062

## BACKGROUND

FREM2 (FRAS1 related extracellular matrix protein 2) is a 3,169 amino acid single-pass type I membrane protein that localizes to the extracellular side of the cell membrane and contains 5 Calx- $\beta$  domains, as well as 12 CSPG repeats. Functioning as an extracellular matrix protein, FREM2 is required for the maintenance of skin and renal epithelia and is also thought to be involved in epidermal adhesion events. Defects or mutations in the gene encoding FREM2, which maps to human chromosome 13q13.3, are associated with Fraser syndrome, a multisystem malformation that is characterized by ear abnormalities, congenital heart defects and cutaneous syndactyly. FREM2 exists as multiple alternatively spliced isoforms.

## REFERENCES

1. Smyth, I., et al. 2004. The extracellular matrix gene Frem1 is essential for the normal adhesion of the embryonic epidermis. *Proc. Natl. Acad. Sci. USA* 101: 13560-13565.
2. Jadeja, S., et al. 2005. Identification of a new gene mutated in Fraser syndrome and mouse myelencephalic blebs. *Nat. Genet.* 37: 520-525.
3. Timmer, J.R., et al. 2005. Tissue morphogenesis and vascular stability require the Frem2 protein, product of the mouse myelencephalic blebs gene. *Proc. Natl. Acad. Sci. USA* 102: 11746-11750.
4. Kiyozumi, D., et al. 2006. Breakdown of the reciprocal stabilization of QBRICK/Frem1, Fras1, and Frem2 at the basement membrane provokes Fraser syndrome-like defects. *Proc. Natl. Acad. Sci. USA* 103: 11981-11986.
5. Shafeghati, Y., et al. 2008. Fraser syndrome due to homozygosity for a splice site mutation of FREM2. *Am. J. Med. Genet. A* 146A: 529-531.
6. van Haelst, M.M., et al. 2008. Molecular study of 33 families with Fraser syndrome new data and mutation review. *Am. J. Med. Genet. A* 146A: 2252-2257.
7. Online Mendelian Inheritance in Man, OMIM™. 2009. Johns Hopkins University, Baltimore, MD. MIM Number: 608945. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>

## CHROMOSOMAL LOCATION

Genetic locus: Frem2 (mouse) mapping to 3 C.

## PRODUCT

FREM2 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 FREM2 shRNA Plasmid (m): sc-75062-SH and FREM2 shRNA (m) Lentiviral Particles: sc-75062-V as alternate gene silencing products.

For independent verification of FREM2 (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-75062A, sc-75062B and sc-75062C.

## RESEARCH USE

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

## 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

FREM2 siRNA (m) is recommended for the inhibition of FREM2 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

FREM2 (F-1): sc-376555 is recommended as a control antibody for monitoring of FREM2 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 FREM2 gene expression knockdown using RT-PCR Primer: FREM2 (m)-PR: sc-75062-PR (20  $\mu$ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## PROTOCOLS

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