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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

Fibromodulin shRNA (m) Lentiviral Particles: sc-44823-V

BACKGROUND

Small leucine-rich proteoglycans (SLRPs) such as Decorin, Biglycan, Fibromodulin, and Lumican mediate extracellular matrix organization and are binding partners of TGF β . Fibromodulin is a collagen-binding Keratan sulphate proteoglycan that influences adhesion processes of connective tissue, and plays a role in fibrillogenesis by regulating collagen fibril spacing and thickness. The core proteins of SLRPs consist of a central region of leucine-rich repeats flanked by disulfide-linkages of the terminal domains. Fibromodulin is a ubiquitous protein that is most prominent in articular cartilage, tendon, and ligament. The human Fibromodulin gene maps to chromosome 1q32 and encodes a 376 amino acid protein.

REFERENCES

1. Antonsson, P., et al. 1993. Structure and deduced amino acid sequence of the human fibromodulin gene. *Biochim. Biophys. Acta* 1174: 204-206.
2. Sztrlovics, R., et al. 1994. Localization of the human fibromodulin gene (FMOD) to chromosome 1q32 and completion of the cDNA sequence. *Genomics* 23: 715-717.
3. Online Mendelian Inheritance in Man, OMIM™. 1995. Johns Hopkins University, Baltimore, MD. MIM Number: 600245. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
4. Roughley, P.J., et al. 1996. Changes with age in the structure of fibromodulin in human articular cartilage. *Osteoarthr. Cartil.* 4: 153-161.
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CHROMOSOMAL LOCATION

Genetic locus: Fmod (mouse) mapping to 1 E4.

PRODUCT

Fibromodulin shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see Fibromodulin siRNA (m): sc-44823 and Fibromodulin shRNA Plasmid (m): sc-44823-SH as alternate gene silencing products.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

APPLICATIONS

Fibromodulin shRNA (m) Lentiviral Particles is recommended for the inhibition of Fibromodulin expression in mouse cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

GENE EXPRESSION MONITORING

Fibromodulin (H-50): sc-33772 is recommended as a control antibody for monitoring of Fibromodulin 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-rabbit IgG-HRP: sc-2004 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible goat anti-rabbit IgG-HRP: sc-2030 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-rabbit IgG-FITC: sc-2012 (dilution range: 1:100-1:400) or goat anti-rabbit IgG-TR: sc-2780 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Fibromodulin gene expression knockdown using RT-PCR Primer: Fibromodulin (m)-PR: sc-44823-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.