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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) 

Fibulin-3 siRNA (h): sc-44624

BACKGROUND

Fibulin-1 is a modular glycoprotein component of elastic extracellular matrix fibers, basement membranes and blood. It can bind calcium, Fibronectin, Laminin, Nidogen and Fibrinogen. Estrogen exposure to ovarian cancer cell lines can upregulate Fibulin-1. Fibulin-2 is abundant in heart, placenta and ovarian tissue and binds several components of the extracellular matrix including aggrecan, versican and brevican. Fibulin-3, also known as EFEMP1, is a secreted protein. Defects in the gene for Fibulin-3 cause the autosomal dominant disease Doyme honeycomb retinal dystrophy (DHRD, also known as malattia leventinese) which is characterized by yellow-white deposits (drusen) that accumulate under the retinal pigment epithelium. Fibulin-3 is not present at the site of drusen formation in normal eyes. Fibulin-5 is an integrin-binding extracellular matrix protein that mediates endothelial cell adhesion.

REFERENCES

1. Ikegawa, S., et al. 1996. Structure and chromosomal assignment of the human S1-5 gene (FBNL) that is highly homologous to fibrillin. *Genomics* 35: 590-592.
2. Stone, E.M., et al. 1999. A single EFEMP1 mutation associated with both malattia leventinese and Doyme honeycomb retinal dystrophy. *Nat. Genet.* 22: 199-202.
3. 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.
4. Matsumoto, M., et al. 2001. Dominant radial drusen and Arg345Trp EFEMP1 mutation. *Am. J. Ophthalmol.* 131: 810-812.
5. Marmorstein, L.Y., et al. 2002. Aberrant accumulation of EFEMP1 underlies drusen formation in malattia leventinese and age-related macular degeneration. *Proc. Natl. Acad. Sci. USA* 99: 13067-13072.
6. Marmorstein, L.Y., et al. 2004. Association of EFEMP1 with malattia leventinese and age-related macular degeneration: a mini-review. *Ophthalmic Genet.* 25: 219-226.

CHROMOSOMAL LOCATION

Genetic locus: EFEMP1 (human) mapping to 2p16.1.

PRODUCT

Fibulin-3 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Fibulin-3 shRNA Plasmid (h): sc-44624-SH and Fibulin-3 shRNA (h) Lentiviral Particles: sc-44624-V as alternate gene silencing products.

For independent verification of Fibulin-3 (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-44624A, sc-44624B and sc-44624C.

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Fibulin-3 siRNA (h) is recommended for the inhibition of Fibulin-3 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 μ M in 66 μ 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-3 (C-3): sc-365224 is recommended as a control antibody for monitoring of Fibulin-3 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 κ BP-HRP: sc-516102 or m-IgG κ 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 κ BP-FITC: sc-516140 or m-IgG κ 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-3 gene expression knockdown using RT-PCR Primer: Fibulin-3 (h)-PR: sc-44624-PR (20 μ 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 for detailed protocols and support products.