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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) 



Profilin-1/2 shRNA (h) Lentiviral Particles: sc-44045-V

BACKGROUND

Profilins regulate Actin polymerization by binding to and sequestering the Actin monomer. Profilins act as a nucleotide exchange factor that charges Actin with ATP after binding the Actin monomer through a 1:1 stoichiometric relationship. Human Profilin-1 and Profilin-2 are encoded by two separate genes mapping to chromosomes 17p13.3 and 3q25.1, respectively. Both Profilin-1 and Profilin-2 are abundantly expressed in kidney. Profilin-1 is highly expressed in lung, liver, placenta and kidney while profilin-2 is highly expressed in brain and skeletal muscle. In axonal and dendritic processes of mouse brain, Profilins co-localize with Dyamin I and Synapsin. Profilin may play a role in mediating cell adhesion. The overexpression of Profilin in endothelial cells results in increased adhesion to Fibronectin. In food allergy, plant Profilin is considered a pan allergen. Case studies indicate individuals with allergies to various foods including celery, carrots, zucchini and peanuts are actually sensitive to the Profilin proteins in these foods.

REFERENCES

1. Kwiatkowski, D.J., et al. 1990. Identification of the functional Profilin gene, its localization to chromosome subband 17p13.3, and demonstration of its deletion in some patients with Miller-Dieker syndrome. *Am. J. Hum. Genet.* 46: 559-567.
2. Goldschmidt-Clermont, P.J., et al. 1992. The control of Actin nucleotide exchange by thymosin b4 and Profilin. A potential regulatory mechanism for Actin polymerization in cells. *Mol. Biol. Cell* 3: 1015-1024.
3. Valenta, R., et al. 1992. Profilins constitute a novel family of functional plant pan-allergens. *J. Exp. Med.* 175: 377-385.
4. Honore, B., et al. 1993. Cloning and expression of a novel human Profilin variant, Profilin-2. *FEBS Lett.* 330: 151-155.
5. Naylor, S.L., et al. 1996. Report of the sixth international workshop on human chromosome 3 mapping 1995. *Cytogenet. Cell Genet.* 72: 255-270.

CHROMOSOMAL LOCATION

Genetic locus: PFN1 (human) mapping to 17p13.2, PFN2 (human) mapping to 3q25.1.

PRODUCT

Profilin-1/2 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 4 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 Profilin-1/2 siRNA (h): sc-44045 and Profilin-1/2 shRNA Plasmid (h): sc-44045-SH as alternate gene silencing products.

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.

APPLICATIONS

Profilin-1/2 shRNA (h) Lentiviral Particles is recommended for the inhibition of Profilin-1/2 expression in human 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

Profilin-1/2 (FL-140): sc-25788 is recommended as a control antibody for monitoring of Profilin-1/2 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 Profilin-1/2 gene expression knockdown using RT-PCR Primer: Profilin-1/2 (h)-PR: sc-44045-PR (20 μ l). Annealing temperature for the primers should be $55-60^\circ\text{C}$ and the extension temperature should be $68-72^\circ\text{C}$.

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.

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.

PROTOCOLS

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