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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) 

periplakin shRNA (m) Lentiviral Particles: sc-43415-V

BACKGROUND

Paraneoplastic pemphigus (PNP) is an autoimmune blistering disease that is associated with underlying neoplasms. PNP sera reacts with multiple plakins family proteins, among which only envoplakin and periplakin are constantly detected. Periplakin, a membrane-associated precursor of the epidermal cornified envelope and desmosomes, is a member of the plakins family of proteins. The periplakin gene PPL is mapped to human chromosome 16p13.3, and its promoter contains multiple AP2 sites and a SP1 site. Periplakin localizes to desmosomes, the interdesmosomal plasma membrane and intermediate filaments. The periplakin rod domain is necessary for the redistribution of envoplakin to desmosomes and the cytoskeleton by heterodimerization. The periplakin linker domain is required for intermediate filament association. The N-terminus of periplakin accumulates at the cell surface of microvilli in association with cortical actin. Envoplakin and periplakin localize independently to desmosomes, and the distribution of envoplakin at the interdesmosomal plasma membrane depends on heterodimerization with periplakin. The N-terminus of periplakin, therefore, plays an important role in forming the scaffold on which the cornified envelope is assembled.

REFERENCES

1. Proby, C., et al. 1999. Human autoantibodies against HD1/plectin in paraneoplastic pemphigus. *J. Invest. Dermatol.* 112: 153-156.
2. Amagai, M. 1999. Autoimmunity against desmosomal cadherins in pemphigus. *J. Dermatol. Sci.* 20: 92-102.
3. Aho, S., et al. 1999. Human periplakin: genomic organization in a clonally unstable region of chromosome 16p with an abundance of repetitive sequence elements. *Genomics* 56: 160-168.

CHROMOSOMAL LOCATION

Genetic locus: Ppl (mouse) mapping to 16 A1.

PRODUCT

periplakin 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 µl 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 periplakin siRNA (m): sc-43415 and periplakin shRNA Plasmid (m): sc-43415-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.

PROTOCOLS

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

APPLICATIONS

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

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl 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

periplakin (C-20): sc-16754 is recommended as a control antibody for monitoring of periplakin 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 donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (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 donkey anti-goat IgG-FITC: sc-2024 (dilution range: 1:100-1:400) or donkey anti-goat IgG-TR: sc-2783 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor periplakin gene expression knockdown using RT-PCR Primer: periplakin (m)-PR: sc-43415-PR (20 µl). 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.

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.