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SANTA CRUZ BIOTECHNOLOGY, INC.

BPAG1 shRNA (h) Lentiviral Particles: sc-43269-V

BACKGROUND

As basal cells of stratified squamous epithelia begin to migrate in response to wound healing, they lose their cell-substrate adhesion junctions, the hemidesmosomes. The hemidesmosome is an adhesion structure of the epidermal-dermal junction in keratinocytes. When keratinocytes migrate laterally or upward to differentiate they must control the formation and disintegration of the hemidesmosomes. The bullous pemphigoid antigen BPAG1 is a hemidesmosomal protein of the cutaneous basement membrane zone. The primary sequence deduced from full-length human cDNAs predicts that this molecule consists of a central rod region and flanking globular domains. A neuronal isoform, BPAG1n3 is the result of differential splicing of BPAG1. BPAG1n3 is distinguished by its initial 32 amino acid residues and by the absence of the amino-terminal half of the Actin-binding domain.

REFERENCES

1. Kitajima, Y., et al. 1992. Phorbol ester- and calcium-induced reorganization of 180-kDa bullous pemphigoid antigen on the ventral surface of cultured human keratinocytes as studied by immunofluorescence and immunoelectron microscopy. Exp. Cell Res. 203: 17-24.
2. Gipson, I.K., et al. 1993. Redistribution of the hemidesmosome components $\alpha 6/\beta 4$ integrin and bullous pemphigoid antigens during epithelial wound healing. Exp. Cell Res. 207: 86-98.
3. Sawamura, D., et al. 1994. Mouse 230-kDa bullous pemphigoid antigen gene: structural and functional characterization of the 5'-flanking region and interspecies conservation of the deduced amino-terminal peptide sequence of the protein. J. Invest. Dermatol. 103: 651-655.
4. Yang, Y., et al. 1999. Integrators of the cytoskeleton that stabilize microtubules. Cell 98: 229-238.

CHROMOSOMAL LOCATION

Genetic locus: DST (human) mapping to 6p12.1.

PRODUCT

BPAG1 shRNA (h) 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 BPAG1 siRNA (h): sc-43269 and BPAG1 shRNA Plasmid (h): sc-43269-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

BPAG1 shRNA (h) Lentiviral Particles is recommended for the inhibition of BPAG1 expression in human 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

BPAG1 (E-14): sc-13776 is recommended as a control antibody for monitoring of BPAG1 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 BPAG1 gene expression knockdown using RT-PCR Primer: BPAG1 (h)-PR: sc-43269-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.