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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

SANTA CRUZ BIOTECHNOLOGY, INC.

BPAG1 siRNA (m): sc-43270

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., Owaribe, K., Nishizawa, Y., Jokura, Y. and Yaoita, H. 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., Spurr-Michaud, S., Tisdale, A., Elwell, J. and Stepp, M.A. 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., Sato, T., Kon, A., Harada, K., Nomura, K., Hashimoto, I., Tamai, K. and Uitto, J. 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. Kitajima, Y., Nojiri, M., Yamada, T., Hirako, Y. and Owaribe, K. 1998. Internalization of the 180 kDa bullous pemphigoid antigen as immune complexes in basal keratinocytes: an important early event in blister formation in bullous pemphigoid. Br. J. Dermatol. 138: 71-76.
5. Yang, Y., Bauer, C., Strasser, G., Wollman, R., Julien, J.P. and Fuchs, E. 1999. Integrators of the cytoskeleton that stabilize microtubules. Cell 98: 229-238.

CHROMOSOMAL LOCATION

Genetic locus: Bpag1 (mouse) mapping to 1 B.

PRODUCT

BPAG1 siRNA (m) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see BPAG1 shRNA Plasmid (m): sc-43270-SH and BPAG1 shRNA (m) Lentiviral Particles: sc-43270-V as alternate gene silencing products.

For independent verification of BPAG1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43270A, sc-43270B and sc-43270C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

BPAG1 siRNA (m) is recommended for the inhibition of BPAG1 expression in mouse 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 µM in 66 µl. 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

BPAG1 (1B10): sc-293499 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 reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 BPAG1 gene expression knockdown using RT-PCR Primer: BPAG1 (m)-PR: sc-43270-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

PROTOCOLS

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