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Diagnostik & molekulare Diagnostik



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DSC1 shRNA (h) Lentiviral Particles: sc-43733-V



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BACKGROUND

The desmosomal cadherin desmocollin DSC1 is expressed in upper epidermis where strong adhesion is required. DSC1 is a type I membrane protein required for strong adhesion and barrier maintenance in epidermis and contributes to epidermal differentiation. DSC3 is also a type I membrane protein and is expressed in all living epidermal layers as well as in glandular ducts, basal matrix cells and the outer root sheath of hair follicles. DSC3, but not DSC1, is also present in desmosomes of the basal and suprabasal cell layers of other stratified epithelia such as cervix, tongue and esophagus as well as in cells of the basal layer of bladder urothelium and the complex epithelium of trachea. The DSC1 gene comprises 17 exons spanning approximately 33 kb on 18q12.1, and the DSC3 gene comprises 17 exons spanning approximately 49 kb on 18q12.1. Alternative splicing gives rise to two DSC1 and DSC3 isoforms, designated 1A and 1B, and 3A and 3B, respectively, which differ in their carboxy termini. DSC1 and DSC3 may contribute to epidermal cell positioning by mediating differential adhesiveness between cells that express different isoforms.

REFERENCES

- Nuber, U.A., et al. 1996. Patterns of desmocollin synthesis in human epithelia: immunolocalization of desmocollins 1 and 3 in special epithelia and in cultured cells. Eur. J. Cell Biol. 71: 1-13.
- Whittock, N.V., et al. 2000. Genomic organization and amplification of the human desmosomal cadherin genes DSC1 and DSC3, encoding desmocollin types 1 and 3. Biochem. Biophys. Res. Commun. 276: 454-460.
- Chidgey, M., et al. 2001. Mice lacking desmocollin 1 show epidermal fragility accompanied by barrier defects and abnormal differentiation. J. Cell Biol. 155: 821-832.
- SWISS-PROT/TrEMBL (GI: 2493423). World Wide Web URL: http://www.expasy.ch/sprot/sprot-top.html

CHROMOSOMAL LOCATION

Genetic locus: DSC1 (human) mapping to 18q12.1.

PRODUCT

DSC1 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 x 10⁶ 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 DSC1 siRNA (h): sc-43733 and DSC1 shRNA Plasmid (h): sc-43733-SH as alternate gene silencing products.

APPLICATIONS

DSC1 shRNA (h) Lentiviral Particles is recommended for the inhibition of DSC1 expression in human cells.

PROTOCOLS

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

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0 x 10⁶ 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

DSC1 (A-4): sc-398590 is recommended as a control antibody for monitoring of DSC1 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-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor DSC1 gene expression knockdown using RT-PCR Primer: DSC1 (h)-PR: sc-43733-PR (20 μ l). 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.

STORAGE

Store lentiviral particles at -80 $^{\circ}$ C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4 $^{\circ}$ C for up to one week. Avoid repeated freeze thaw cycles.

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.

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