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desmoplakin I/II siRNA (h): sc-43724

BACKGROUND

Desmosomes are major cell adhesion junctions that are particularly prominent in the epidermis and in cardiac tissue and are important for the rigidity and strength of the cell. The desmosome consists of several proteins, of which desmoplakin is the most abundant. Desmoplakin plays an important role in the attachment of the filaments to the desmosome. Specifically, desmoplakin interacts with plakophilin 1 (PKP1), PKP2 or PKP3, or combinations thereof, to selectively recruit plakophilins to desmosomal plaques. Desmoplakin has also been shown to function as a transglutaminase substrate *in vitro*, suggesting that it may participate in cell adhesion at the intraepidermal level. Desmoplakin exists as a two-stranded coiled coil structure. Alternative splicing gives rise to two isoforms, desmoplakin I and II, which differ by 600 amino acids.

REFERENCES

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2. Norgett, E.E., Hatsell, S.J., Carvajal-Huerta, L., Cabezas, J.C., Common, J., Purkis, P.E., Whittock, N., Leigh, I.M., Stevens, H.P. and Kelsell, D.P. 2000. Recessive mutation in desmoplakin disrupts desmoplakin-intermediate filament interactions and causes dilated cardiomyopathy, woolly hair and keratoderma. *Hum. Mol. Genet.* 9: 2761-2766.
3. Hofmann, I., Mertens, C., Brette, I. M., Nimmrich, V., Schnolzer, M. and Herrmann, H. 2000. Interaction of plakophilins with desmoplakin and intermediate filament proteins: an *in vitro* analysis. *J. Cell Sci.* 113: 2471-2483.
4. Esposito, C., Lombardi, M.L., Ruocco, V., Cozzolino, A., Marinello, L. and Porta, R. 2000. Implication of tissue transglutaminase and desmoplakin in cell adhesion mechanism in human epidermis. *Mol. Cell. Biochem.* 206: 57-65.

CHROMOSOMAL LOCATION

Genetic locus: DSP (human) mapping to 6p24.3.

PRODUCT

desmoplakin I/II siRNA (h) is a pool of 4 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see desmoplakin I/II shRNA Plasmid (h): sc-43724-SH and desmoplakin I/II shRNA (h) Lentiviral Particles: sc-43724-V as alternate gene silencing products.

For independent verification of desmoplakin I/II (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-43724A, sc-43724B, sc-43724C and sc-43724D.

PROTOCOLS

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

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 μ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330 μ l of RNase-free water makes a 10 μ M solution in a 10 μ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

desmoplakin I/II siRNA (h) is recommended for the inhibition of desmoplakin I/II expression in human 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 μ M in 66 μ l. 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

desmoplakin I/II (A-1): sc-390975 is recommended as a control antibody for monitoring of desmoplakin I/II 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 desmoplakin I/II gene expression knockdown using RT-PCR Primer: desmoplakin I/II (h)-PR: sc-43724-PR (20 μ l, 572 bp). 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.