

vezatin siRNA (h): sc-43199

BACKGROUND

Vezatin is a single transmembrane domain containing mammalian adhesion protein that is ubiquitously expressed at adherens cell-cell junctions. Adherens junctions (zonula adherens) are cell-cell junctions that host microfilaments and/or intermediate filaments, which can coordinate with focal adhesion proteins and mediate tissue organization and morphogenesis. Vezatin interacts with actin filamentous networks and anchors myosin VIIA to cadherin complexes, thereby creating a network between adherens junctions and the actin cytoskeleton. This may enhance cell-cell adhesion characteristics and influence cadherin-based signals. Vezatin is concentrated in the fibrillar links interconnecting the bases of adjacent stereocilia in the inner ear sensory hair cells and may mediate proper positioning of hair cell stereocilia. Loss of a functional vezatin-myosin VIIA complex at both the adherent junctions and the base of the stereocilia is likely to account for the splaying out of the stereocilia observed in *Myo7a*^{-/-} animals. Vezatin recruitment to adherens junctions implicates the C-terminal region of α -catenin.

REFERENCES

1. Kemler, R. 1993. From cadherins to catenins: cytoplasmic protein interactions and regulation of cell adhesion. *Trends Genet.* 9: 317-321.
2. Kussel-Andermann, P., et al. 2000. Vezatin, a novel transmembrane protein, bridges Myosin VIIA to the cadherin-catenins complex. *EMBO J.* 19: 6020-6029.
3. Ko, K.S., et al. 2001. Cadherins mediate intercellular mechanical signaling in fibroblasts by activation of stretch-sensitive calcium permeable channels. *J. Biol. Chem.* 276: 35967-35977.
4. Li, G., et al. 2001. N-cadherin-mediated intercellular interactions promote survival and migration of melanoma cells. *Cancer Res.* 61: 3819-3825.
5. Geisbrecht, E.R. and Montell D.J. 2002. Myosin VI is required for E-cadherin-mediated border cell migration. *Nat. Cell Biol.* 4: 616-620.
6. Blaschuk, O.W., et al. 2002. Plasma membrane components of adherens junctions (Review). *Mol. Membr. Biol.* 19: 75-80.
7. Sousa, S., et al. 2004. Unconventional Myosin VIIA and vezatin, two proteins crucial for *Listeria* entry into epithelial cells. *J. Cell Sci.* 117: 2121-2130.

CHROMOSOMAL LOCATION

Genetic locus: VEZT (human) mapping to 12q22.

PRODUCT

vezatin siRNA (h) 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see vezatin shRNA Plasmid (h): sc-43199-SH and vezatin shRNA (h) Lentiviral Particles: sc-43199-V as alternate gene silencing products.

For independent verification of vezatin (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-43199A, sc-43199B and sc-43199C.

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

vezatin siRNA (h) is recommended for the inhibition of vezatin 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

vezatin (B-1): sc-271347 is recommended as a control antibody for monitoring of vezatin 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 vezatin gene expression knockdown using RT-PCR Primer: vezatin (h)-PR: sc-43199-PR (20 μ l). 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.