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Diagnostik & molekulare Diagnostik



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Fascin 3 shRNA (m) Lentiviral Particles: sc-44620-V



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BACKGROUND

Cell adhesion to extracellular matrix is an important physiological stimulus for organization of the Actin-based cytoskeleton. Adhesion to the matrix gly-coprotein thrombospondin-1 triggers the sustained formation of F-Actin microspikes that contain the Actin-bundling protein fascin. These structures are also implicated in cell migration, which may be an important function of throm-bospondin-1 in tissue remodelling and wound repair. Fascin bundles Actin microfilaments within dynamic cellular structures such as microspikes, stress fibers and membrane ruffles. Fascin could serve as a prognostic factor for abnormal ovarian epithelial pathology and could be a novel target for the treatment of ovarian cancer. Fascin, an Actin-bundling protein, identifies dendritic cells in the blood and in tissues. Fascin 3 (FSCN3) is primarily expressed in testis.

REFERENCES

- 1. Jaffe, R., et al. 1998. Fascin and the differential diagnosis of childhood histiocytic lesions. Pediatr. Dev. Pathol. 1: 216-221.
- 2. Adams, J.C., et al. 2000. Stimulation of fascin spikes by thrombospondin-1 is mediated by the GTPases Rac and Cdc42. J. Cell Biol. 150: 807-822.
- Tubb, B.E., et al. 2000. Characterization of human retinal fascin gene (FSCN2) at 17q25: close physical linkage of fascin and cytoplasmic actin genes. Genomics 65: 146-156.
- 4. Hu, W., et al. 2000. Increased expression of fascin, motility associated protein, in cell cultures derived from ovarian cancer and in borderline and carcinomatous ovarian tumors. Clin. Exp. Metastasis 18: 83-88.
- Grothey, A., et al. 2000. Fascin, an actin-bundling protein associated with cell motility, is upregulated in hormone receptor negative breast cancer.
 Br. J. Cancer 83: 870-873.
- Tubb, B., et al. 2002. Testis fascin (FSCN3): a novel paralog of the actinbundling protein fascin expressed specifically in the elongate spermatid head. Exp. Cell Res. 275: 92-109.

CHROMOSOMAL LOCATION

Genetic locus: Fscn3 (mouse) mapping to 6 A3.3.

PRODUCT

Fascin 3 shRNA (m) 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 μ l frozen stock containing 1.0 x 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 Fascin 3 siRNA (m): sc-44620 and Fascin 3 shRNA Plasmid (m): sc-44620-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.

APPLICATIONS

Fascin 3 shRNA (m) Lentiviral Particles is recommended for the inhibition of Fascin 3 expression in mouse cells.

SUPPORT REAGENTS

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

Fascin 3 (D-15): sc-32607 is recommended as a control antibody for monitoring of Fascin 3 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 Fascin 3 gene expression knockdown using RT-PCR Primer: Fascin 3 (m)-PR: sc-44620-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.

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.

PROTOCOLS

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