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Vav3 shRNA (h) Lentiviral Particles: sc-44187-V

BACKGROUND

The Vav family of Rho guanine nucleotide exchange factors (GEFs) orchestrate signaling events following lymphocyte antigen receptor activation. Vav3, like Vav (also known as Vav1 or p95Vav), undergoes tyrosine phosphorylation downstream of T cell receptor cross-linkage, and subsequently interacts with two adaptor molecules, SLP76 and 3BP2. Following these events, however, the paths of Vav and Vav3 diverge; Vav affects IL-2 promoter activity, while Vav3 impacts gene transcription linked to serum response element (SRE). Furthermore, Vav3 expression follows a cell cycle-dependent pattern, with transient upregulation occurring during mitosis. Enforced Vav3 expression leads to the appearance of multinucleate cells, implicating a role for Vav3 in the control of cytokinesis.

REFERENCES

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CHROMOSOMAL LOCATION

Genetic locus: VAV3 (human) mapping to 1p13.3.

PRODUCT

Vav3 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 μ l frozen stock containing 1.0×10^6 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 Vav3 siRNA (h): sc-44187 and Vav3 shRNA Plasmid (h): sc-44187-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

Vav3 shRNA (h) Lentiviral Particles is recommended for the inhibition of Vav3 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 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

Vav3 (K-19): sc-28474 is recommended as a control antibody for monitoring of Vav3 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 Vav3 gene expression knockdown using RT-PCR Primer: Vav3 (h)-PR: sc-44187-PR (20 μ l, 585 bp). Annealing temperature for the primers should be $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ 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.

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