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Rap 2B shRNA (h) Lentiviral Particles: sc-43515-V

BACKGROUND

Ras oncogenes encode GTP-binding proteins that are capable of transforming immortalized cells in culture. Two Ras-related human genes, designated RAP1A and RAP1B, encode 95% homologous proteins (namely Rap 1A and Rap 1B) that share a similar C-terminal Cys-Ali-Ali-Xaa sequence with Ras proteins and are ubiquitously expressed in mammalian tissues. The putative "effector" domain of Ras proteins, whose integrity is required for cell transformation as well as interaction with the putative effector protein GAP, is conserved in both Rap 1 proteins. Rap 1A is thought to interfere with Ras effector function by binding to Ras GAP in a GTP-dependent manner without affecting Rap 1A GTPase activity. Rap 2, another Ras-related protein, shares 60% identity with Rap 1A and exhibits a carboxy terminal CAAX motif and two upstream cysteines similar to those of the H-Ras, K-Ras and N-Ras proteins. In contrast with Rap 1A and Rap 1B, overexpression of Rap 2 does not interfere with the Ras signaling pathway.

REFERENCES

1. Pizon, V., et al. 1988. Human cDNAs Rap 1 and Rap 2 homologous to the *Drosophila* gene Dras3 encode proteins closely related to ras in the "effector" region. *Oncogene* 3: 201-204.
2. Pizon, V., et al. 1988. Nucleotide sequence of a human cDNA encoding a Ras-related protein (rap1B). *Nucleic Acids Res.* 16: 7719.
3. Culine, S., et al. 1989. Expression of the Ras-related rap genes in human tumors. *Int. J. Cancer* 44: 990-994.
4. Kitayama, H., et al. 1989. A Ras-related gene with transformation suppressor activity. *Cell* 56: 77-84.
5. Frech, M., et al. 1990. Inhibition of GTPase activating protein stimulation of Ras-p21 GTPase by the Krev-1 gene product. *Science* 249: 169-171.

CHROMOSOMAL LOCATION

Genetic locus: RAP2B (human) mapping to 3q25.2.

PRODUCT

Rap 2B 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 1.0×10^6 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 Rap 2B siRNA (h): sc-43515 and Rap 2B shRNA Plasmid (h): sc-43515-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.

PROTOCOLS

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

APPLICATIONS

Rap 2B shRNA (h) Lentiviral Particles is recommended for the inhibition of Rab 2B expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rab 2B gene expression knockdown using RT-PCR Primer: Rap 2B (h)-PR: sc-43515-PR (20 µl). Annealing temperature for the primers should be $55-60^{\circ}\text{C}$ and the extension temperature should be $68-72^{\circ}\text{C}$.

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.