Rab 9A siRNA (h): sc-41830



The Power to Question

BACKGROUND

The Ras-related superfamily of guanine nucleotide binding proteins includes the R-Ras, Rap, Ral/Rec and Rho/Rab subfamilies all of which are thought to play an important role in either endocytosis or in biosynthetic protein transport. The process of transporting newly synthesized proteins from the endoplasmic reticulum (ER) to various stacks of the Golgi complex and to secretory vesicles involves the movement of carrier vesicles and requires Rab protein function. Rab proteins are also an integral part of endocytic pathways. Rab 9A is a 201 amino acid protein that localizes to the cytoplasmic side of the cell membrane, as well as to the membrane of the Golgi apparatus and the ER, and is involved in the transport of proteins between endosomes and the *trans* Golgi network.

REFERENCES

- Davies, J.P., et al. 1997. Cloning and mapping of human Rab7 and Rab9 cDNA sequences and identification of a Rab9 pseudogene. Genomics 41: 131-134.
- Díaz, E., et al. 1997. A novel Rab9 effector required for endosome-to-TGN transport. J. Cell Biol. 138: 283-290.
- 3. de Leeuw, H.P., et al. 1998. Small GTP-binding proteins in human endothelial cells. Br. J. Haematol. 103: 15-19.
- Seki, N., et al. 2000. cDNA cloning of a new member of the Ras superfamily, RAB9-like, on the human chromosome Xq22.1-q22.3 region. J. Hum. Genet. 45: 318-322.
- 5. Carroll, K.S., et al. 2001. Role of Rab9 GTPase in facilitating receptor recruitment by TIP47. Science 292: 1373-1376.
- Barbero, P., et al. 2002. Visualization of Rab9-mediated vesicle transport from endosomes to the *trans*-Golgi in living cells. J. Cell Biol. 156: 511-518.
- 7. Walter, M., et al. 2003. Telomerase immortalization upregulates Rab9 expression and restores LDL cholesterol egress from Niemann-Pick C1 late endosomes. J. Lipid Res. 44: 243-253.
- 8. Ganley, I.G., et al. 2004. Rab9 GTPase regulates late endosome size and requires effector interaction for its stability. Mol. Biol. Cell 15: 5420-5430.
- 9. Aivazian, D., et al. 2006. TIP47 is a key effector for Rab9 localization. J. Cell Biol. 173: 917-926.

CHROMOSOMAL LOCATION

Genetic locus: RAB9A (human) mapping to Xp22.2.

PRODUCT

Rab 9A siRNA (h) is a target-specific 19-25 nt siRNA 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 Rab 9A shRNA Plasmid (h): sc-41830-SH and Rab 9A shRNA (h) Lentiviral Particles: sc-41830-V as alternate gene silencing products.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

Rab 9A siRNA (h) is recommended for the inhibition of Rab 9A 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 µM in 66 µl. 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

Rab 9A (Mab9): sc-53145 is recommended as a control antibody for monitoring of Rab 9A 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 Rab 9A gene expression knockdown using RT-PCR Primer: Rab 9A (h)-PR: sc-41830-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.