

Rab GAP1 siRNA (m): sc-152654

BACKGROUND

GTPase-activating proteins (GAPs) accelerate the intrinsic rate of GTP hydrolysis of Ras-related proteins, resulting in downregulation of their active form. Rab GAP1 (RAB GTPase activating protein 1), also known as GAPCEN (GAP and centrosome-associated) or TBC1D11, is a 1,069 amino acid cytoplasmic protein containing one PID domain and one Rab-GAP TBC domain. Rab GAP1 acts as a GTPase-activating protein of Rab 6A in the Rab 6A-mediated pathway, which is involved in the metaphase-anaphase transition. Expressed as four alternatively spliced variants, Rab GAP1 may also be involved in the microtubule nucleation by centrosome and in Golgi dynamics during the cell cycle. Rab GAP1 is encoded by a gene located on human chromosome 9q33.2, which consists of about 145 million bases, 4% of the human genome and encodes nearly 900 genes.

REFERENCES

1. Cuif, M.H., Possmayer, F., Zander, H., Bordes, N., Jollivet, F., Couedel-Courteille, A., Janoueix-Lerosey, I., Langsley, G., Bornens, M. and Goud, B. 1999. Characterization of GAPCenA, a GTPase activating protein for Rab6, part of which associates with the centrosome. *EMBO J.* 18: 1772-1782.
2. Echard, A., Opdam, F.J., de Leeuw, H.J., Jollivet, F., Savelkoul, P., Hendriks, W., Voorberg, J., Goud, B. and Fransen, J.A. 2000. Alternative splicing of the human Rab6A gene generates two close but functionally different isoforms. *Mol. Biol. Cell* 11: 3819-3833.
3. Miserey-Lenkei, S., Couedel-Courteille, A., Del Nery, E., Bardin, S., Piel, M., Racine, V., Sibarita, J.B., Perez, F., Bornens, M. and Goud, B. 2006. A role for the Rab6A GTPase in the inactivation of the Mad2-spindle checkpoint. *EMBO J.* 25: 278-289.
4. Yoshimura, S., Egerer, J., Fuchs, E., Haas, A.K. and Barr, F.A. 2007. Functional dissection of Rab GTPases involved in primary cilium formation. *J. Cell Biol.* 178: 363-369.
5. Caratù, G., Allegra, D., Bimonte, M., Schiattarella, G.G., D'Ambrosio, C., Scaloni, A., Napolitano, M., Russo, T. and Zambrano, N. 2007. Identification of the ligands of protein interaction domains through a functional approach. *Mol. Cell. Proteomics* 6: 333-345.
6. Yoshimura, S., Haas, A.K. and Barr, F.A. 2008. Analysis of Rab GTPase and GTPase-activating protein function at primary cilia. *Meth. Enzymol.* 439: 353-364.
7. Kanno, E., Ishibashi, K., Kobayashi, H., Matsui, T., Ohbayashi, N. and Fukuda, M. 2010. Comprehensive screening for novel rab-binding proteins by GST pull-down assay using 60 different mammalian Rabs. *Traffic* 11: 491-507.

CHROMOSOMAL LOCATION

Genetic locus: Rabgap1 (mouse) mapping to 2 B.

PROTOCOLS

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

PRODUCT

Rab GAP1 siRNA (m) 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 Rab GAP1 shRNA Plasmid (m): sc-152654-SH and Rab GAP1 shRNA (m) Lentiviral Particles: sc-152654-V as alternate gene silencing products.

For independent verification of Rab GAP1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-152654A, sc-152654B and sc-152654C.

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

Rab GAP1 siRNA (m) is recommended for the inhibition of Rab GAP1 expression in mouse 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Rab GAP1 gene expression knockdown using RT-PCR Primer: Rab GAP1 (m)-PR: sc-152654-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.