

ARHGAP1 siRNA (h): sc-96477

BACKGROUND

GTPase-activating proteins (GAPs) accelerate the intrinsic rate of GTP hydrolysis of Ras-related proteins, resulting in downregulation of their active form. ARHGAP1 (Rho GTPase activating protein 1), also known as CDC42GAP or RhoGAP1, is a 439 amino acid protein that localizes to the cytoplasm and contains one Rho-GAP domain and one CRAL-TRIO domain. Expressed ubiquitously, ARHGAP1 exists in a complex with several other proteins, including eIF4A1 and Exportin 7, and functions as a GTPase activator for Rho, Rac and Cdc42 proteins, effectively converting them to an inactive GDP-bound state. The gene encoding ARHGAP1 maps to human chromosome 11, which houses over 1,400 genes and comprises nearly 4% of the human genome. Jervell and Lange-Nielsen syndrome, Jacobsen syndrome, Niemann-Pick disease, hereditary angioedema and Smith-Lemli-Opitz syndrome are associated with defects in genes that maps to chromosome 11.

REFERENCES

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2. Diekmann, D., et al. 1991. Bcr encodes a GTPase-activating protein for p21rac. *Nature* 351: 400-402.
3. Barfod, E.T., et al. 1993. Cloning and expression of a human CDC42 GTPase-activating protein reveals a functional SH3-binding domain. *J. Biol. Chem.* 268: 26059-26062.
4. Lancaster, C.A., et al. 1994. Characterization of rhoGAP. A GTPase-activating protein for rho-related small GTPases. *J. Biol. Chem.* 269: 1137-1142.
5. Zhou, Y.T., et al. 2005. BNIP-2 induces cell elongation and membrane protrusions by interacting with Cdc42 via a unique Cdc42-binding motif within its BNIP-2 and Cdc42GAP homology domain. *Exp. Cell Res.* 303: 263-274.
6. Wildenberg, G.A., et al. 2006. p120-catenin and p190RhoGAP regulate cell-cell adhesion by coordinating antagonism between Rac and Rho. *Cell* 127: 1027-1039.
7. Engelse, M.A., et al. 2008. Differential gene expression analysis of tubule forming and non-tubule forming endothelial cells: CDC42GAP as a counter-regulator in tubule formation. *Angiogenesis* 11: 153-167.

CHROMOSOMAL LOCATION

Genetic locus: ARHGAP1 (human) mapping to 11p11.2.

PRODUCT

ARHGAP1 siRNA (h) 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 ARHGAP1 shRNA Plasmid (h): sc-96477-SH and ARHGAP1 shRNA (h) Lentiviral Particles: sc-96477-V as alternate gene silencing products.

For independent verification of ARHGAP1 (h) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-96477A, sc-96477B and sc-96477C.

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

ARHGAP1 siRNA (h) is recommended for the inhibition of ARHGAP1 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 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.

GENE EXPRESSION MONITORING

ARHGAP1 (C-10): sc-398671 is recommended as a control antibody for monitoring of ARHGAP1 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 goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (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 goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor ARHGAP1 gene expression knockdown using RT-PCR Primer: ARHGAP1 (h)-PR: sc-96477-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.