SANTA CRUZ BIOTECHNOLOGY, INC.

Rho GDIα siRNA (h): sc-36417

BACKGROUND

Members of the Ras superfamily of small GTP-binding proteins are critical mediators of diverse cell signaling pathways, including those leading to cell proliferation, cytoskeletal organization and secretion. The counter-conversion of the active GTP-bound form of these proteins to their inactive GDP-bound form is influenced by two types of regulatory proteins: those that alter the intrinsic GTPase activity of the GTP-binding proteins and those that alter the rate of GDP/GTP exchange. Guanine nucleotide-releasing factors (GRFs) increase the GDP dissociation rate, while GDP-dissociation inhibitors (GDIs) decrease the dissociation rate. Rho GDIα, also known as ARHGDIA or GDIA1, is a 204 amino acid member of the Rho GDI family of proteins. Localized to the cytoplasm, Rho GDIα inhibits the dissociation of GDP from Rho proteins, thereby preventing GTP from binding to and subsequently activating Rho proteins. In humans, Rho GDIα can be phosphorylated at Ser 101 by p21-activated kinase (αPAK), an event that inhibits Rho GDIα activity and may result in positive feedback regulation of certain Rho GDIα target proteins.

REFERENCES

1. Leffers, H., et al. 1993. Identification of two human Rho GDP dissociation inhibitor proteins whose overexpression leads to disruption of the Actin cytoskeleton. Exp. Cell Res. 209: 165-174.
2. Wagner, T., et al. 1997. A somatic cell hybrid panel for distal 17q: GDIA1 maps to 17q25.3. Cytogenet. Cell Genet. 76: 172-175.
3. Di-Poï, N., et al. 2001. Mechanism of NADPH oxidase activation by the Rac/Rho-GDI complex. Biochemistry 40: 10014-10022.

CHROMOSOMAL LOCATION

Genetic locus: ARHGDIA (human) mapping to 17q25.3.

PRODUCT

Rho GDIα 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Rho GDIα shRNA Plasmid (h): sc-36417-SH and Rho GDIα shRNA (h) Lentiviral Particles: sc-36417-V as alternate gene silencing products.

For independent verification of Rho GDIα (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-36417A, sc-36417B and sc-36417C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Rho GDIα siRNA (h) is recommended for the inhibition of Rho GDIα 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

Rho GDIα (G-2): sc-373724 is recommended as a control antibody for monitoring of Rho GDIα 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-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, UltraCruz® Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 Rho GDIα gene expression knockdown using RT-PCR Primer: Rho GDIα (h)-PR: sc-36417-PR (20 µl, 349 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Rong, F., et al. 2012. Knockdown of Rho GDIα induces apoptosis and increases lung cancer cell chemosensitivity to paclitaxel. Neoplasma 59: 541-550.

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.