

Rac 1 siRNA (h2): sc-44325

BACKGROUND

A large number of low molecular weight, GTP binding proteins of the Ras superfamily have been identified. These proteins regulate many fundamental processes in all eukaryotic cells such as growth, vesicle traffic and cytoskeletal organization. GTPase-activating proteins (GAPs) accelerate the intrinsic rate of GTP hydrolysis of Ras-related proteins, resulting in downregulation of their active form. Two proteins in this family, Rac 1 and Rac 2, are 92% identical and share GTP binding and GTP hydrolysis motifs with other members of the Ras superfamily. Rac 1 is expressed in a large number of different cell types. Rac 2 is primarily expressed only in myeloid cells and has been reported to be a regulatory component of the human neutrophil NADPH oxidase.

REFERENCES

1. Trahey, M. and McCormick, F. 1987. A cytoplasmic protein stimulates normal N-Ras p21 GTPase, but does not affect oncogenic mutants. *Science* 238: 542-545.
2. Sewell, J.L. and Kahn, R.A. 1988. Sequences of the bovine and yeast ADP-ribosylation factor and comparison to other GTP-binding proteins. *Proc. Natl. Acad. Sci. USA* 85: 4620-4624.
3. Didsbury, J., et al. 1989. Rac, a novel Ras-related family of proteins that are botulinum toxin substrates. *J. Biol. Chem.* 264: 16378-16382.
4. Hall, A. 1990. The cellular functions of small GTP-binding proteins. *Science* 249: 636-640.
5. Xu, G., et al. 1990. The catalytic domain of the neurofibromatosis type 1 gene product stimulates ras GTPase and complements ira mutants of *S. cerevisiae*. *Cell* 63: 835-841.

CHROMOSOMAL LOCATION

Genetic locus: RAC1 (human) mapping to 7p22.1.

PRODUCT

Rac 1 siRNA (h2) 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 Rac 1 shRNA Plasmid (h2): sc-44325-SH and Rac 1 shRNA (h2) Lentiviral Particles: sc-44325-V as alternate gene silencing products.

For independent verification of Rac 1 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44325A, sc-44325B and sc-44325C.

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

Rac 1 siRNA (h2) is recommended for the inhibition of Rac 1 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

pan Rac (G-2): sc-514583 is recommended as a control antibody for monitoring of Rac 1 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 Rac 1 gene expression knockdown using RT-PCR Primer: Rac 1 (h2)-PR: sc-44325-PR (20 μ l, 443 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Shields, M.A., et al. 2012. Interplay between β 1-integrin and Rho signaling regulates differential scattering and motility of pancreatic cancer cells by snail and slug proteins. *J. Biol. Chem.* 287: 6218-6229.
2. Jamieson, C., et al. 2015. Rac1 augments Wnt signaling by stimulating β -catenin-lymphoid enhancer factor-1 complex assembly independent of β -catenin nuclear import. *J. Cell Sci.* 128: 3933-3946.

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.