

RGNEF siRNA (h): sc-92013

BACKGROUND

The Ras superfamily of GTPases can be subdivided into the Ras, Rho/Rac, Sar, Rab, Arf, Rap and Ran subfamilies, all of which control multiple aspects of cell function, including cytoskeletal rearrangement, nuclear signaling and cell growth. The Ras superfamily of GTPases function as regulated switches that toggle between a biologically active GTP-bound and an inactive GDP-bound form. This activation is catalyzed by guanine nucleotide exchange factors (GEFs). RGNEF (Rho-guanine nucleotide exchange factor), also known as p190RhoGEF, is a 1,705 amino acid protein that functions as a Rho A-specific GEF, regulating signaling pathways downstream of growth factor receptors (GFR) and Integrins. Localized to both the cytoplasm and the cell membrane, RGNEF exists as a homooligomer that, in addition to functioning as a Rho A-specific GEF, plays an important role in cell motility, apoptosis and dendritic morphogenesis. Four isoforms of RGNEF exist due to alternative splicing events.

REFERENCES

- Meyer, D., et al. 1999. Interaction of c-Jun amino-terminal kinase interacting protein-1 with p190 rhoGEF and its localization in differentiated neurons. *J. Biol. Chem.* 274: 35113-35118.
- Zhai, J., et al. 2001. Identification of a novel interaction of 14-3-3 with p190RhoGEF. *J. Biol. Chem.* 276: 41318-41324.
- van Horck, F.P., et al. 2001. Characterization of p190RhoGEF, a RhoA-specific guanine nucleotide exchange factor that interacts with microtubules. *J. Biol. Chem.* 276: 4948-4956.
- Ge, W., et al. 2002. Binding of p190RhoGEF to a destabilizing element on the light neurofilament mRNA is competed by BC1 RNA. *J. Biol. Chem.* 277: 42701-42705.
- Wu, J., et al. 2003. Cytoplasmic retention sites in p190RhoGEF confer anti-apoptotic activity to an EGFP-tagged protein. *Brain Res. Mol. Brain Res.* 117: 27-38.
- Zhai, J., et al. 2003. Direct interaction of focal adhesion kinase with p190RhoGEF. *J. Biol. Chem.* 278: 24865-24873.
- Hemsath, L., et al. 2005. Fluorescence approaches for monitoring interactions of Rho GTPases with nucleotides, regulators, and effectors. *Methods* 37: 173-182.
- Kim, H., et al. 2008. δ -catenin-induced dendritic morphogenesis. An essential role of p190RhoGEF interaction through Akt1-mediated phosphorylation. *J. Biol. Chem.* 283: 977-987.
- Lim, Y., et al. 2008. PyK2 and FAK connections to p190Rho guanine nucleotide exchange factor regulate RhoA activity, focal adhesion formation, and cell motility. *J. Cell Biol.* 180: 187-203.

CHROMOSOMAL LOCATION

Genetic locus: ARHGEF28 (human) mapping to 5q13.2.

PROTOCOLS

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

PRODUCT

RGNEF 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 RGNEF shRNA Plasmid (h): sc-92013-SH and RGNEF shRNA (h) Lentiviral Particles: sc-92013-V as alternate gene silencing products.

For independent verification of RGNEF (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-92013A, sc-92013B and sc-92013C.

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

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

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor RGNEF gene expression knockdown using RT-PCR Primer: RGNEF (h)-PR: sc-92013-PR (20 μ l, 597 bp). 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.