

SGEF siRNA (h): sc-78233

BACKGROUND

The Ras superfamily of GTPases, which can be subdivided into the Ras, Rho/Rac, Sar, Rab, ARF and Ran subfamilies, controls 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). SGEF (SH3-containing guanine nucleotide exchange factor), also known as CSGEF, is an 871 amino acid RhoGEF that is highly expressed in adult liver tissue and is thought to regulate membrane dynamics during macropinocytosis (invagination of the cell membrane to form a vesicle pocket). SGEF is found in prostate cancer cells, suggesting a role for SGEF in carcinogenesis. Multiple isoforms of SGEF exist due to alternative splicing events.

REFERENCES

1. Qi, H., et al. 2003. Isolation of the novel human guanine nucleotide exchange factor Src homology 3 domain-containing guanine nucleotide exchange factor (SGEF) and of C-terminal SGEF, an N-terminally truncated form of SGEF, the expression of which is regulated by androgen in prostate cancer cells. *Endocrinology* 144: 1742-1752.
2. Chong, Y.P., et al. 2004. A novel non-catalytic mechanism employed by the C-terminal Src-homologous kinase to inhibit Src-family kinase activity. *J. Biol. Chem.* 279: 20752-20766.
3. Ellerbroek, S.M., et al. 2004. SGEF, a Rho G guanine nucleotide exchange factor that stimulates macropinocytosis. *Mol. Biol. Cell* 15: 3309-3319.
4. McGee, A.W., et al. 2004. Calcium channel function regulated by the SH3-GK module in β subunits. *Neuron* 42: 89-99.
5. Patel, J.C. and Galán, J.E. 2006. Differential activation and function of Rho GTPases during Salmonella-host cell interactions. *J. Cell Biol.* 175: 453-463.

CHROMOSOMAL LOCATION

Genetic locus: ARHGEF26 (human) mapping to 3q25.2.

PRODUCT

SGEF 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 SGEF shRNA Plasmid (h): sc-78233-SH and SGEF shRNA (h) Lentiviral Particles: sc-78233-V as alternate gene silencing products.

For independent verification of SGEF (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-78233A, sc-78233B and sc-78233C.

PROTOCOLS

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

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

SGEF siRNA (h) is recommended for the inhibition of SGEF 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

SGEF (E-5): sc-514048 is recommended as a control antibody for monitoring of SGEF 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 SGEF gene expression knockdown using RT-PCR Primer: SGEF (h)-PR: sc-78233-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.