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SRGAP1 siRNA (m): sc-153820

BACKGROUND

SRGAPs contain a highly conserved overall primary structure and play an important role in the cell facilitating Slit- robo signaling in cell migration and axon guidance. SRGAP1 (Slit- robo Rho GTPase activating protein 1), also known as ARHGAP13 (Rho GTPase activating protein 13), functions as a GTPase-activating protein for Cdc42 and Rho A. Expressed in kidney, testis, lung and brain, SRGAP1 contains an FCH (Fes/CIP4 homology) domain, a Rho-GAP domain and an SH3 domain. In the presence of Slit, SRGAP1 (via its SH3 domain) binds to the CC3 motif in robo (a protein responsible for mediating the repulsive effect of Slit) with higher affinity and inhibits Cdc42 activity in a robo/SRGAP-dependent manner. More specifically, SRGAP1 increases the intrinsic GTPase activity of Cdc42, thereby converting it to its inactive, GDP-bound form. Inactivation of Cdc42 ultimately leads to a decrease in Actin polymerization.

REFERENCES

1. Nagase, T., et al. 2000. Prediction of the coding sequences of unidentified human genes. XVI. The complete sequences of 150 new cDNA clones from brain which code for large proteins *in vitro*. DNA Res. 7: 65-73.
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4. Katoh, M. and Katoh, M. 2003. FNBP2 gene on human chromosome 1q32.1 encodes ARHGAP family protein with FCH, FBH, RhoGAP and SH3 domains. Int. J. Mol. Med. 11: 791-797.
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6. Katoh, Y. and Katoh, M. 2004. Identification and characterization of ARHGAP27 gene in silico. Int. J. Mol. Med. 14: 943-947.
7. Katoh, M. and Katoh, M. 2004. Characterization of human ARHGAP10 gene in silico. Int. J. Oncol. 25: 1201-1206.

CHROMOSOMAL LOCATION

Genetic locus: Srgap1 (mouse) mapping to 10 D2.

PRODUCT

SRGAP1 siRNA (m) 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 SRGAP1 shRNA Plasmid (m): sc-153820-SH and SRGAP1 shRNA (m) Lentiviral Particles: sc-153820-V as alternate gene silencing products.

For independent verification of SRGAP1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-153820A, sc-153820B and sc-153820C.

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

SRGAP1 siRNA (m) is recommended for the inhibition of SRGAP1 expression in mouse 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

SRGAP1 (D-11): sc-390349 is recommended as a control antibody for monitoring of SRGAP1 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 SRGAP1 gene expression knockdown using RT-PCR Primer: SRGAP1 (m)-PR: sc-153820-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.

PROTOCOLS

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