SANTA CRUZ BIOTECHNOLOGY, INC.

ARHGAP6 siRNA (h): sc-91170

BACKGROUND

ARHGAP6 (Rho GTPase activating protein 6), also known as RHOGAP6 or RHOGAPX-1, is a widely expressed cytoplasmic protein that functions as a GTPase-activating protein specific for RhoA and as a cytoskeletal protein promoting actin remodeling. ARHGAP6 contains three SH3 binding domains and a Rho GAP domain. In the cell, ARHGAP6 colocalizes with actin filaments and, via its N-terminus, recruits F-Actin. The gene encoding ARHGAP6 is located on the X chromosome in the region that is deleted in patients with microphthalmia with linear skin defects (MLS), an X-linked dominant male-lethal syndrome. MLS is characterized by aplastic skin, agenesis of the corpus callosum and microphthalmia (a condition in which eyes fail to enlarge due to a malformation of the choroid fissure). Despite its absence in MLS patients, ARHGAP6 does not appear to participate in the pathogenesis of the disease.

REFERENCES

1. Tribioli, C., et al. 1996. An X chromosome-linked gene encoding a protein with characteristics of a rhoGAP predominantly expressed in hematopoietic cells. Proc. Natl. Acad. Sci. USA 93: 695-699.
2. Schaefer, L., et al. 1997. Cloning and characterization of a novel Rho-type GTPase-activating protein gene (ARHGAP6) from the critical region for microphthalmia with linear skin defects. Genomics 46: 268-277.
3. Schmidt, A., et al. 1999. Ectopic expression of RET results in microphthalmia and tumors in the retinal pigment epithelium. Int. J. Cancer 80: 600-605.
4. Prakash, S.K., et al. 2000. Functional analysis of ARHGAP6, a novel GTPase-activating protein for RhoA. Hum. Mol. Genet. 9: 477-488.
5. Kutsche, K., et al. 2002. Microphthalmia with linear skin defects syndrome (MLS): a male with a mosaic paracentric inversion of Xp. Cytogenet. Genome Res. 99: 297-302.
6. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 300118. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: ARHGAP6 (human) mapping to Xp22.2.

PRODUCT

ARHGAP6 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 ARHGAP6 shRNA Plasmid (h): sc-91170-SH and ARHGAP6 shRNA (h) Lentiviral Particles: sc-91170-V as alternate gene silencing products.

For independent verification of ARHGAP6 (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-91170A, sc-91170B and sc-91170C.

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

ARHGAP6 siRNA (h) is recommended for the inhibition of ARHGAP6 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

ARHGAP6 (D-5): sc-81938 is recommended as a control antibody for monitoring of ARHGAP6 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 ARHGAP6 gene expression knockdown using RT-PCR Primer: ARHGAP6 (h)-PR: sc-91170-PR (20 µl). 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.