

ARAP3 siRNA (h): sc-61988

BACKGROUND

The ADP-ribosylation factor (ARF) family of small GTP-binding proteins are involved in vesicular transport regulation and in controlling cytoskeletal organization and cell adhesion. These proteins are best characterized as regulators of membrane traffic. The Centaurin GTPase-activating protein family comprise a subset of ARF regulatory molecules that transduce PI 3-kinase activation into coordinated control of ARF-dependent pathways. ARAP3 (ankyrin repeat and Pleckstrin homology domain-containing protein 3), also known as Centaurin- δ 3 (Cnt- δ 3), acts as a GTPase activating protein for ARF and Rho G proteins. ARAP3 consists of a Ras association (RA) domain, five Pleckstrin homology (PH) domains, three ankyrin repeats, a sterile α motif (SAM) domain, a Rho-GAP domain and an Arf-GAP domain. ARAP3 localizes to the cytoplasm and is ubiquitously expressed, with highest expression in peripheral blood leukocytes.

REFERENCES

1. Krugmann, S., et al. 2002. Identification of ARAP3, a novel PI3K effector regulating both Arf and Rho GTPases, by selective capture on phosphoinositide affinity matrices. *Mol. Cell* 9: 95-108.
2. Lu, Q., et al. 2004. EST-based genome-wide gene inactivation identifies ARAP3 as a host protein affecting cellular susceptibility to anthrax toxin. *Proc. Natl. Acad. Sci. USA* 101: 17246-17251.
3. I, S.T., et al. 2004. ARAP3 is transiently tyrosine phosphorylated in cells attaching to Fibronectin and inhibits cell spreading in a RhoGAP-dependent manner. *J. Cell Sci.* 117: 6071-6084.
4. Krugmann, S., et al. 2004. ARAP3 is a PI3K- and RAP-regulated GAP for RhoA. *Curr. Biol.* 14: 1380-1384.
5. Logan, M.R. and Mandato, C.A. 2006. Regulation of the Actin cytoskeleton by PIP2 in cytokinesis. *Biol. Cell* 98: 377-388.
6. Krugmann, S., et al. 2006. ARAP3 is essential for formation of lamellipodia after growth factor stimulation. *J. Cell Sci.* 119: 425-432.
7. Lindmo, K. and Stenmark, H. 2006. Regulation of membrane traffic by phosphoinositide 3-kinases. *J. Cell Sci.* 119: 605-614.
8. Krugmann, S., et al. 2006. Purification of ARAP3 and characterization of GAP activities. *Meth. Enzymol.* 406: 91-103.

CHROMOSOMAL LOCATION

Genetic locus: ARAP3 (human) mapping to 5q31.3.

PRODUCT

ARAP3 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 ARAP3 shRNA Plasmid (h): sc-61988-SH and ARAP3 shRNA (h) Lentiviral Particles: sc-61988-V as alternate gene silencing products.

For independent verification of ARAP3 (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-61988A, sc-61988B and sc-61988C.

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

ARAP3 siRNA (h) is recommended for the inhibition of ARAP3 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 ARAP3 gene expression knockdown using RT-PCR Primer: ARAP3 (h)-PR: sc-61988-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Tang, Z., et al. 2012. Exchange protein activated by cyclic adenosine monophosphate regulates the switch between adipogenesis and osteogenesis of human mesenchymal stem cells through increasing the activation of phosphatidylinositol 3-kinase. *Int. J. Biochem. Cell Biol.* 44: 1106-1120.
2. Daubon, T., et al. 2016. VEGF-A stimulates podosome-mediated collagen-IV proteolysis in microvascular endothelial cells. *J. Cell Sci.* 129: 2586-2598.

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.