

# AGAP1 siRNA (h): sc-44443

## BACKGROUND

ARFGAP with GTP-binding protein-like, Ankyrin repeat and pleckstrin homology domains 1 (AGAP1), also designated Centaurin  $\gamma$  2 (CENTG2), is a member of the ADP-ribosylation factor family of ARF6 GTPase-activating proteins (GAP). GAPs are important regulators of ARF function by controlling ARFs return to its inactive state. AGAP1, which is endosome-associated and phosphoinositide-dependent, regulates the adapter protein 3 (AP-3)-dependent trafficking of proteins in the endosomal-lysosomal system. The protein associates with the endocytic compartment in the cytoplasm and has an effect on the Actin cytoskeleton. Overexpression of AGAP1 induces a loss of Actin stress fibers. AGAP1 is related to ACAP1 and ASAP1, and all three proteins are similarly expressed in fibroblast cells such as NIH/3T3.

## REFERENCES

1. Nie, Z., et al. 2002. AGAP1, an endosome-associated, phosphoinositide-dependent ADP-ribosylation factor, GTPase-activating protein that affects Actin cytoskeleton. *J. Biol. Chem.* 277: 48965-48975.
2. Nie, Z., et al. 2003. Specific regulation of the adaptor protein complex AP-3 by the ARFGAP AGAP1. *Dev. Cell* 5: 513-521.
3. Meurer, S., et al. 2004. AGAP1, a novel binding partner of nitric oxide-sensitive guanylyl cyclase. *J. Biol. Chem.* 279: 49346-49354.
4. Che, M.M., et al. 2005. Assays and properties of the ARFGAPs AGAP1, ASAP1 and ARFGAP1. *Methods Enzymol.* 404: 147-163.
5. Nie, Z., et al. 2005. The ARFGAPs AGAP1 and AGAP2 distinguish between the adaptor protein complexes AP-1 and AP-3. *J. Cell Sci.* 118: 3555-3566.
6. Wassink, T.H., et al. 2005. Evaluation of the chromosome 2q37.3 gene CENTG2 as an autism susceptibility gene. *Am. J. Med. Genet. B Neuropsychiatr. Genet.* 136: 36-44.
7. SWISS-PROT/TrEMBL (Q9UPQ3). World Wide Web URL: <http://www.expasy.ch/sprot/sprot-top.html>

## CHROMOSOMAL LOCATION

Genetic locus: AGAP1 (human) mapping to 2q37.2.

## PRODUCT

AGAP1 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see AGAP1 shRNA Plasmid (h): sc-44443-SH and AGAP1 shRNA (h) Lentiviral Particles: sc-44443-V as alternate gene silencing products.

For independent verification of AGAP1 (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-44443A, sc-44443B and sc-44443C.

## PROTOCOLS

See our web site at [www.scbt.com](http://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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

AGAP1 siRNA (h) is recommended for the inhibition of AGAP1 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  $\mu$ M in 66  $\mu$ 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 AGAP1 gene expression knockdown using RT-PCR Primer: AGAP1 (h)-PR: sc-44443-PR (20  $\mu$ 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.