



## RASA4 siRNA (h): sc-89880

### BACKGROUND

RASA4 (ras GTPase-activating protein 4) is an 803 amino acid member of the GAP1 family of GTPase-activating proteins that suppresses the Ras/mitogen-activated protein kinase pathway in response to  $Ca^{2+}$ . Stimuli that increase intracellular  $Ca^{2+}$  levels result in the translocation of the RASA4 protein to the plasma membrane, where it activates Ras GTPase activity. Consequently, Ras is converted from the active GTP-bound state to the inactive GDP-bound state and no longer activates downstream pathways that regulate gene expression, cell growth and differentiation. Containing 21 exons and spanning 35 kb of genomic DNA, RASA4 includes one Btk-type zinc finger, 2 C2 domains, one PH domain and one Ras-GAP domain. Existing as two alternatively spliced isoforms, the RASA4 gene is conserved in canine, bovine, mouse, chicken and zebrafish, and maps to human chromosome 7q22.1.

### REFERENCES

1. Minagawa, T., et al. 2001. Distinct phosphoinositide binding specificity of the GAP1 family proteins: characterization of the pleckstrin homology domains of MRASAL and KIAA0538. *Biochem. Biophys. Res. Commun.* 288: 87-90.
2. Lockyer, P.J., et al. 2001. CAPRI regulates  $Ca^{2+}$ -dependent inactivation of the Ras-MAPK pathway. *Curr. Biol.* 11: 981-986.
3. Hillier, L.W., et al. 2003. The DNA sequence of human chromosome 7. *Nature* 424: 157-164.
4. Bivona, T.G., et al. 2003. Phospholipase C $\gamma$  activates Ras on the Golgi apparatus by means of RasGRP1. *Nature* 424: 694-698.
5. Online Mendelian Inheritance in Man, OMIM<sup>TM</sup>. 2003. Johns Hopkins University, Baltimore, MD. MIM Number: 607943. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
6. Stelzl, U., et al. 2005. CAPRI and RASAL impose different modes of information processing on Ras due to contrasting temporal filtering of  $Ca^{2+}$ . *J. Cell Biol.* 170: 183-190.

### CHROMOSOMAL LOCATION

Genetic locus: RASA4 (human) mapping to 7q22.1.

### PRODUCT

RASA4 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 RASA4 shRNA Plasmid (h): sc-89880-SH and RASA4 shRNA (h) Lentiviral Particles: sc-89880-V as alternate gene silencing products.

For independent verification of RASA4 (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-89880A, sc-89880B and sc-89880C.

### 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  $-20^{\circ}$  C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at  $-20^{\circ}$  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

RASA4 siRNA (h) is recommended for the inhibition of RASA4 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 RASA4 gene expression knockdown using RT-PCR Primer: RASA4 (h)-PR: sc-89880-PR (20  $\mu$ l). Annealing temperature for the primers should be  $55-60^{\circ}$  C and the extension temperature should be  $68-72^{\circ}$  C.

### RESEARCH USE

For research use only, not for use in diagnostic procedures.