SANTA CRUZ BIOTECHNOLOGY, INC.

# RAPGEF6 siRNA (m): sc-76350

## BACKGROUND

RAPGEF6 (Rap guanine nucleotide exchange factor 6), also known as PDZGEF2 (PDZ domain-containing guanine nucleotide exchange factor 2) or RA-GEF-2, is a guanine nucleotide exchange factor (GEF) that is expressed in a variety of tissues. Localizing to the cytoplasm and translocated to the plasma membrane upon ligand binding, RAPGEF6 contains an N-terminal Ras-GEF domain, a cyclic nucleotide monophosphate-binding domain, a PDZ (PSD-95/DlgA/ZO-1) domain, a Ras-associating (RA) domain and a Ras exchanger motif. RAPGEF6 is closely related to RAPGEF2 and both proteins exhibit GEF activity specific towards Rap 1 and Rap 2. In addition, RAPGEF6 is capable of binding to M-Ras via its RA domain. Due to alternative splicing events, two additional isoforms exist for RAPGEF6, namely PDZ-GEF2A and PDZ-GEF2B.

## REFERENCES


1. Gao, X., et al. 2001. Identification and characterization of RA-GEF-2, a Rap guanine nucleotide exchange factor that serves as a downstream target of M-Ras. J. Biol. Chem. 276: 42219-4222
2. Kozlov, G., et al. 2002. Solution structure of the PDZ2 domain from cytosolic human phosphatase hPTP1E complexed with a peptide reveals contribution of the β2-β3 loop to PDZ domain-ligand interactions. J. Mol. Biol. 320: 813-820.
3. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 610499. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Papp, R., et al. 2003. ESI-MS and FTIR studies of the interaction between the second PDZ domain of hPTP1E and target peptides. Biochem. Cell Biol. 81: 71-80.
5. Kuiperij, H.B., et al. 2003. Characterisation of PDZ-GEFs, a family of guanine nucleotide exchange factors specific for Rap1 and Rap2. Biochim. Biophys. Acta 1593: 141-149.


## CHROMOSOMAL LOCATION

Genetic locus: Rapgef6 (mouse) mapping to 11 B1.3.

## PRODUCT

RAPGEF6 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see RAPGEF6 shRNA Plasmid (m): sc-76350-SH and RAPGEF6 shRNA (m) Lentiviral Particles: sc-76350-V as alternate gene silencing products.

For independent verification of RAPGEF6 (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-76350A, sc-76350B and sc-76350C.

## PROTOCOLS

See our web site at 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 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

RAPGEF6 siRNA (m) is recommended for the inhibition of RAPGEF6 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 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

RAPGEF6 (F-8): sc-398642 is recommended as a control antibody for monitoring of RAPGEF6 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 RAPGEF6 gene expression knockdown using RT-PCR Primer: RAPGEF6 (m)-PR: sc-76350-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.