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### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

# PDZ-RhoGEF siRNA (m): sc-45824

## BACKGROUND

The multidomain (RGS)-containing RhoGEFs represent a family of guanine nucleotide exchange factors that stabilize the nucleotide-free state of small GTPases through their DH/PH domains, leading to the exchange of GDP to GTP. Uniquely, PDZ-RhoGEF, also known as Rho guanine nucleotide exchange factor 11 and ARHGEF11, binds tightly to both nucleotide-free and activated Rho A, therefore playing a role as a primary regulator of Rho A. Mutations within the carboxylate-binding loop of PDZ-RhoGEF result in changes in cell morphology and actin organization which is likely due to its interaction with MAP-1A (MAP1 light chain LC2). PDZ-RhoGEF also plays a role in B plexin-mediated activation of Rho/Rho kinase signaling, which is implicated in the regulation of axon guidance and cell migration.

## REFERENCES

1. Fukuhara, S., et al. 1999. A novel PDZ domain containing guanine nucleotide exchange factor links heterotrimeric G proteins to Rho. *J. Biol. Chem.* 274: 5868-5879.
2. Rumenapp, U., et al. 1999. Rho-specific binding and guanine nucleotide exchange catalysis by KIAA0380, a DBL family member. *FEBS. Lett.* 459: 313-318.
3. Garrard, S.M., et al. 2001. Expression, purification, and crystallization of the RGS-like domain from the Rho nucleotide exchange factor, PDZ-RhoGEF, using the surface entropy reduction approach. *Protein. Expr. Purif.* 21: 412-416.
4. Driessens, M.H., et al. 2002. B plexins activate Rho through PDZ-RhoGEF. *FEBS. Lett.* 529: 168-172.
5. Oleksy, A., et al. 2004. Preliminary crystallographic analysis of the complex of the human GTPase RhoA with the DH/PH tandem of PDZ-RhoGEF. *Acta. Crystallogr. D. Biol. Crystallogr.* 60: 740-742.
6. Tanabe, S., et al. 2004. Regulation of RGS-RhoGEFs by G $_{\alpha 12}$  and G $_{\alpha 13}$  proteins. *Methods Enzymol.* 390: 285-294.

## CHROMOSOMAL LOCATION

Genetic locus: Arhgef11 (mouse) mapping to 3 F1.

## PRODUCT

PDZ-RhoGEF 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PDZ-RhoGEF shRNA Plasmid (m): sc-45824-SH and PDZ-RhoGEF shRNA (m) Lentiviral Particles: sc-45824-V as alternate gene silencing products.

For independent verification of PDZ-RhoGEF (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-45824A, sc-45824B and sc-45824C.

## 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

PDZ-RhoGEF siRNA (m) is recommended for the inhibition of PDZ-RhoGEF 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  $\mu$ M in 60  $\mu$ 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.

## GENE EXPRESSION MONITORING

Ox40L (RR19): sc-80271 is recommended as a control antibody for monitoring of Ox40L (h) 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).

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PDZ-RhoGEF gene expression knockdown using RT-PCR Primer: PDZ-RhoGEF (m)-PR: sc-45824-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.