

PLC δ siRNA (m): sc-155939

BACKGROUND

Phosphoinositide-specific phospholipase C (PLC) plays a crucial role in the initiation of receptor-mediated signal transduction through the generation of the two second messengers, inositol 1,4,5-triphosphate (IP₃) and diacylglycerol (DAG) from phosphatidylinositol 4,5-bisphosphate. There are several mammalian PLC proteins, including PLC β 1, PLC β 2, PLC β 3, PLC β 4, PLC γ 1, PLC γ 2, PLC δ 1, PLC δ 3, PLC δ 4 and PLC ϵ . PLC δ 1, a calcium signal amplifier, is activated by an atypical GTP-binding protein and functions as an effector for GTP-binding protein transglutaminase II-mediated oxytocin receptor and α_{1B} -adrenoreceptor signaling. PLC δ 1 is highly expressed in brain, heart, lung and testis and is abnormally accumulated in autopsied brains with Alzheimer's disease (AD), suggesting that it may play a role in the pathology of AD. Both PLC δ 3 and PLC δ 4 contain several functional domains through which they bind calcium as a cofactor and catalyze the creation of DAG and IP₃, playing an essential role in signal transduction. PLC δ 4 is highly expressed in skeletal muscle and kidney tissue, as well as in corneal epithelial cells, suggesting a role in the regulation of kidney and ocular function.

REFERENCES

1. Ghosh, S., et al. 1997. Phospholipase C isoforms δ 1 and δ 3 from human fibroblasts. High-yield expression in *Escherichia coli*, simple purification, and properties. *Protein Expr. Purif.* 9: 262-278.
2. Pawelczyk, T., et al. 1997. Expression, purification and kinetic properties of human recombinant phospholipase C δ 3. *Acta Biochim. Pol.* 44: 221-229.
3. Kim, H., et al. 1999. Assignment of the human PLC δ 3 gene (PLCD3) to human chromosome band 17q21 by fluorescence *in situ* hybridization. *Cytogenet. Cell Genet.* 87: 209-210.
4. Pawelczyk, T., et al. 1999. Phospholipase C- δ 3 binds with high specificity to phosphatidylinositol 4,5-bisphosphate and phosphatidic acid in bilayer membranes. *Eur. J. Biochem.* 262: 291-298.
5. Lin, F.G., et al. 2001. Downregulation of phospholipase C δ 3 by cAMP and calcium. *Biochem. Biophys. Res. Commun.* 286: 274-280.
6. Ananthanarayanan, B., et al. 2002. Membrane targeting of C2 domains of phospholipase C- δ isoforms. *J. Biol. Chem.* 277: 3568-3575.

CHROMOSOMAL LOCATION

Genetic locus: Plcd3 (mouse) mapping to 11 E1.

PRODUCT

PLC δ 3 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PLC δ 3 shRNA Plasmid (m): sc-155939-SH and PLC δ 3 shRNA (m) Lentiviral Particles: sc-155939-V as alternate gene silencing products.

For independent verification of PLC δ 3 (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-155939A, sc-155939B and sc-155939C.

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

PLC δ 3 siRNA (m) is recommended for the inhibition of PLC δ 3 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 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.

GENE EXPRESSION MONITORING

PLC δ 3 (E-5): sc-514912 is recommended as a control antibody for monitoring of PLC δ 3 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-IgG κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG κ 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 PLC δ 3 gene expression knockdown using RT-PCR Primer: PLC δ 3 (m)-PR: sc-155939-PR (20 μ 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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.