SANTA CRUZ BIOTECHNOLOGY, INC.

PLC β1 siRNA (r): sc-270424

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The Power to Question

BACKGROUND

Phosphoinositide-specific phospholipase C (PLC) plays a critical role in the initiation of receptor mediated signal transduction through the generation of the two second messengers, inositol 1,4,5-triphosphate and diacylglycerol from phosphatidylinositol 4,5 bisphosphate. There are many mammalian PLC isozymes, including PLC β1, PLC β2, PLC β3, PLC β4, PLC γ1, PLC γ2, PLC δ1 and PLC δ2 and PLCε. PLC β1, one of the PLC β isozymes, exists as two immunologically distinguishable proteins (PLC β1a) and (PLC β1b). The two isoforms encode in two distinct transcripts and are generated by alternative splicing of a single gene. PLC β1a is preferentially expressed in the cytosol, whereas PLC β1b is predominantly localized in the nuclei. PLC β1 is a G protein-dependent phosphodiesterase that hydrolyses phosphatidylinositol 4,5 biphosphate into inositol 1,4,5-triphosphate and diacylglycerol after the stimulation of a variety of neurotransmitter receptors at the cell surface. The C-terminal region of PLC β1 has G_q GAP activity and has ability to interact with G_q and other PLC β1 molecules.

REFERENCES

1. Suh, P., et al. 1988. Inositol phospholipid-specific phospholipase C: complete cDNA and protein sequences and sequence homology to tyrosine kinase-related oncogene products. Proc. Natl. Acad. Sci. USA 85: 5419-5423.
2. Emori, Y., et al. 1989. A second type of rat phosphoinositide-specific phospholipase C containing a Src-related sequence not essential for phosphoinositide-hydrolyzing activity. J. Biol. Chem. 264: 21885-21890.
3. Meldrum, E., et al. 1991. A second gene product of the inositol-phospholipid-specific phospholipase Cδ subclass. Eur. J. Biochem. 196: 159-165.

CHROMOSOMAL LOCATION

Genetic locus: Plcb1 (rat) mapping to 3q36.

PRODUCT

PLC β1 siRNA (r) 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 PLC β1 shRNA Plasmid (r): sc-270424-SH and PLC β1 shRNA (r) Lentiviral Particles: sc-270424-V as alternate gene silencing products.

For independent verification of PLC β1 (r) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-270424A, sc-270424B and sc-270424C.

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

PLC β1 siRNA (r) is recommended for the inhibition of PLC β1 expression in rat 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

PLC β1 (D-8): sc-5291 is recommended as a control antibody for monitoring of PLC β1 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 PLC β1 gene expression knockdown using RT-PCR Primer: PLC β1 (r)-PR: sc-270424-PR (20 µl, 482 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Atef, M.E. and Anand-Srivastava, M.B. 2014. Enhanced expression of $G_{q\,\alpha}$ and PLC-β1 proteins contributes to vascular smooth muscle cell hypertrophy in SHR: role of endogenous Angiotensin II and endothelin-1. Am. J. Physiol., Cell Physiol. 307: C97-C106.

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.