SANTA CRUZ BIOTECHNOLOGY, INC.

PLC β4 siRNA (h): sc-36274

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. A total of eight mammalian PLC isozymes have been described (PLC β1, PLC β2, PLC β3, PLC β4, PLC γ1, PLC γ2, PLC δ1 and PLC δ2). The γ-type enzymes are unique in that they contain SH2 and SH3 domains. Moreover, the two γ-type enzymes, but not the β and δ isozymes, are subject to activation by a number of protein tyrosine kinases which associate with their SH2 domains and induce their activation by phosphoryation. In contrast, activation of PLC β1, PLC β2 and PLC β3 is mediated by the a subunits of the G_q class of heterotrimeric G proteins and by certain bg G protein subunits. The regulatory mechanisms for PLC δ1 and PLC δ2 are not yet resolved.

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.
4. Koch, C.A., et al. 1991. SH2 and SH3 domains: elements that control interactions of cytoplasmic signaling proteins. Science 252: 668-674.

CHROMOSOMAL LOCATION

Genetic locus: PLCB4 (human) mapping to 20p12.3.

PRODUCT

PLC β4 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PLC β4 shRNA Plasmid (h): sc-36274-SH and PLC β4 shRNA (h) Lentiviral Particles: sc-36274-V as alternate gene silencing products.

For independent verification of PLC β4 (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-36274A, sc-36274B and sc-36274C.

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 β4 siRNA (h) is recommended for the inhibition of PLC β4 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 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 β4 (A-8): sc-166131 is recommended as a control antibody for monitoring of PLC β4 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 β4 gene expression knockdown using RT-PCR Primer: PLC β4 (h)-PR: sc-36274-PR (20 µl, 597 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Hong, J., et al. 2010. Bile acid reflux contributes to development of esoph-ageal adenocarcinoma via activation of phosphatidylinositol-specific phospholipase C γ2 and NADPH oxidase NOX5-S. Cancer Res. 70: 1247-1255.

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.