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PLC β 3 siRNA (h2): sc-44321

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 β 1, PLC β 2, PLC β 3, PLC β 4, PLC γ 1, PLC γ 2, PLC δ 1 and PLC δ 2). 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 phosphorylation. In contrast, activation of PLC β 1, PLC β 2 and PLC β 3 is mediated by the α subunits of the G_q class of heterotrimeric G proteins and by certain $\beta\gamma$ G protein subunits. The regulatory mechanisms for PLC δ 1 and PLC δ 2 are as yet not resolved.

REFERENCES

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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.
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5. Rhee, S.G. and Choi, K.D. 1992. Regulation of inositol phospholipid-specific phospholipase C isozymes. *J. Biol. Chem.* 267: 12393-12396.
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CHROMOSOMAL LOCATION

Genetic locus: Plcb3 (mouse) mapping to 19 A.

PRODUCT

PLC β 3 siRNA (h2) 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 β 3 shRNA Plasmid (h2): sc-44321-SH and PLC β 3 shRNA (h2) Lentiviral Particles: sc-44321-V as alternate gene silencing products.

For independent verification of PLC β 3 (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-44321A, sc-44321B and sc-44321C.

PROTOCOLS

See our web site at www.scbt.com or our catalog 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 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 β 3 siRNA (h2) is recommended for the inhibition of PLC β 3 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 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 β 3 (H-1): sc-271372 is recommended as a control antibody for monitoring of 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor gene expression knockdown using RT-PCR Primer: PLC β 3 (h2)-PR: sc-44321-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.