SANTA CRUZ BIOTECHNOLOGY, INC.

PC-PLD1 siRNA (h): sc-44000

BACKGROUND

Virtually every cell uses phosphatidylcholine as a substrate to produce phosphatidic acid and choline. Phosphatidylcholine phospholipase D1 and D2 (PC-PLD1 and PC-PLD2) are phospholipid-specific phosphodiesterases that hy-drolyze phosphatidylcholine. Unlike PC-PLD1, which associates with secretory granules, PC-PLD2 localizes to the plasma membrane, where it is implicated in the formation of endocytotic vesicles. Both PC-PLD1 and PC-PLD2 coordinately regulate macrophage phagocytosis. PC-PLD activity in mammalian cells is transiently stimulated upon activation by G protein-coupled and receptor tyrosine kinase cell surface receptors. For example, PC-PLD1 and PC-PLD2 participate in sphingosine 1-phosphate stimulation of ERK phosphorylation and IL-8 secretion in bronchial epithelial cells. In addition, tubulin binding to PC-PLD2 inhibits muscarinic receptor-linked PC-PLD2 activation. PC-PLD2 also enhances PKCζ activity through direct interaction in a lipase activity-independent manner. PC-PLD1 and PC-PLD2 stimulate cell growth by repressing expression of p21 gene through p53-dependent and p53-independent pathways, respectively, which may ultimately lead to carcinogenesis.

CHROMOSOMAL LOCATION

Genetic locus: PLD1 (human) mapping to 3q26.31.

PRODUCT

PC-PLD1 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 PC-PLD1 shRNA Plasmid (h): sc-44000-SH and PC-PLD1 shRNA (h) Lentiviral Particles: sc-44000-V as alternate gene silencing products.

For independent verification of PC-PLD1 (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-44000A, sc-44000B and sc-44000C.

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

PC-PLD1 siRNA (h) is recommended for the inhibition of PC-PLD1 expression in human cells.

PROTOCOLS

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

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

PC-PLD1 (F-12): sc-28314 is recommended as a control antibody for monitoring of PC-PLD1 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 PC-PLD1 gene expression knockdown using RT-PCR Primer: PC-PLD1 (h)-PR: sc-44000-PR (20 µl, 437 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Usatyuk, P.V., et al. 2009. Phospholipase D-mediated activation of IQGAP1 through Rac1 regulates hyperoxia-induced $p47^{phox}$ translocation and reactive oxygen species generation in lung endothelial cells. J. Biol. Chem. 284: 15339-15352.
2. Chen, F., et al. 2013. Phospholipase D2 mediates signaling by ATPase class I type 8B membrane 1. J. Lipid Res. 54: 379-385.
3. Abdulnour, R.E., et al. 2018. Phospholipase D isoforms differentially regulate leukocyte responses to acute lung injury. J. Leukoc. Biol. 103: 919-932.
4. Zhang, G., et al. 2020. $CD4^+$ T cell-mimicking nanoparticles broadly neutralize HIV-1 and suppress viral replication through autophagy. mBio 11: e00903-e00920.
5. Stricker, H.M., et al. 2021. The phospholipase D inhibitor FIPI potently blocks EGF-induced calcium signaling in human breast cancer cells. Cell Commun. Signal. 19: 43.
6. Mahmud, S., et al. 2023. Lipopolysaccharide stimulates A549 cell migration through p-Tyr 42 RhoA and phospholipase D1 activity. Biomolecules 14: 6.

RESEARCH USE

For research use only, not for use in diagnostic procedures.