SANTA CRUZ BIOTECHNOLOGY, INC.

$cPLA_2$ siRNA (h): sc-29280

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

Phospholipase A_2s (PLA_2s) constitute a family of esterases that hydrolyze the sn-2-acyl ester bond in glycerophospholipid molecules. These enzymes are generally calcium-dependent and have been found both intra- and extracellularly. By hydrolyzing the sn-2 bond in glycerophospholipids, PLA_2s release fatty acids. One such fatty acid, arachidonic acid, generates the substrates for the initiation of the arachidonic acid cascade that produces various eicosanoids (i.e. prostaglandins, leukotrienes and thromboxanes), many of which are potent mediators of inflammation. PLA_2s include both the relatively low molecular weight type I and type II enzymes and the form known as cytoplasmic PLA_2 ($cPLA_2$). $cPLA_2$ is present in the cytosol of various cells and tissues including platelets, macrophages and monoblasts; and preferentially hydrolyzes the sn-2 position of phospholipid molecules, releasing free arachidonate.

CHROMOSOMAL LOCATION

Genetic locus: PLA2G4A (human) mapping to 1q31.1.

PRODUCT

$cPLA_2$ siRNA (h) is a target-specific 19-25 nt siRNA 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 $cPLA_2$ shRNA Plasmid (h): sc-29280-SH and $cPLA_2$ shRNA (h) Lentiviral Particles: sc-29280-V as alternate gene silencing 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 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

$cPLA_2$ siRNA (h) is recommended for the inhibition of $cPLA_2$ 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.

RESEARCH USE

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

GENE EXPRESSION MONITORING

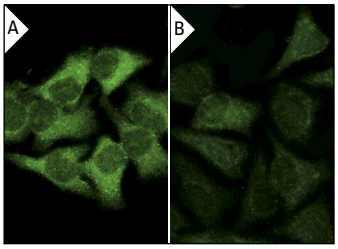
$cPLA_2$ (4-4B-3C): sc-454 is recommended as a control antibody for monitoring of $cPLA_2$ gene expression knockdown by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) or immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500).

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 $cPLA_2$ gene expression knockdown using RT-PCR Primer: $cPLA_2$ (h)-PR: sc-29280-PR (20 µl, 466 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

DATA



$cPLA_2$ siRNA (h): sc-29280. Immunofluorescence staining of methanol-fixed, control HeLa (**A**) and $cPLA_2$ siRNA silenced HeLa (**B**) cells showing diminished cytoplasmic staining in the siRNA silenced cells. Cells probed with $cPLA_2$ (C-20): sc-1724.

SELECT PRODUCT CITATIONS

1. Anwar, K., et al. 2011. COX-2 inhibition and inhibition of cytosolic Phospholipase A_2 increase CD36 expression and foam cell formation in THP-1 cells. Lipids 46: 131-142.
2. Tang, F., et al. 2013. Interleukin 15 primes natural killer cells to kill via NKG2D and $cPLA_2$ and this pathway is active in psoriatic arthritis. PLoS ONE 8: e76292.

PROTOCOLS

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