group X sPLA₂ siRNA (m): sc-60031



The Power to Question

BACKGROUND

Phospholipases catalyze the release of fatty acids from phospho-lipids. One member of the phospholipase family, iPLA $_2$, is detected as a membrane-bound protein with multiple smaller isoforms, which result from alternative splicing. Another phopholipase, sPLA $_2$, belongs to a family of secretory phospholipases A $_2$, which represent an expanding family of related enzymes. sPLA $_2$ has both membrane bound and secreted forms that are encoded by a single gene which maps to human chromosome 1p35. sPLA $_2$ is involved in the regulation of phospholipid metabolism in biomembranes and in eicosanoid biosynthesis. group X sPLA $_2$ mRNA is found in various tissues including the lung, thymus, and spleen, and immunohistochemical analysis reveals its expression in splenic macrophages. group X sPLA $_2$ is an actively secreted enzyme that maps to human chromosome 16p13.1-p12.

REFERENCES

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- Cupillard, L., et al. 1997. Cloning, chromosomal mapping, and expression of a novel human secretory phospholipase A₂. J. Biol. Chem. 272: 15745-15752.
- 4. Kitadokoro, K., et al. 1998. Crystal structure of human secretory phospholipase A_2 -IIA complex with the potent indolizine inhibitor 120-1032. J. Biochem. 123: 619-623.
- 5. Ma, Z., et al. 1999. Human pancreatic islets express mRNA species encoding two distinct catalytically active isoforms of group VI phospholipase A_2 (iPLA₂) that arise from an exon-skipping mechanism of alternative splicing of the transcript from the iPLA₂ gene on chromosome 22q13.1. J. Biol. Chem. 274: 9607-9616.
- 6. Morioka, Y., et al. 2000. Mouse group X secretory phospholipase A₂ induces a potent release of arachidonic acid from spleen cells and acts as a ligand for the phospholipase A₂ receptor. Arch. Biochem. Biophys. 381: 31-42.

CHROMOSOMAL LOCATION

Genetic locus: Pla2g10 (mouse) mapping to 16 A1.

PRODUCT

group X sPLA $_2$ siRNA (m) 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 group X sPLA $_2$ shRNA Plasmid (m): sc-60031-SH and group X sPLA $_2$ shRNA (m) Lentiviral Particles: sc-60031-V as alternate gene silencing products.

For independent verification of group X $\rm sPLA_2$ (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: $\rm sc-60031A$, $\rm sc-60031B$ and $\rm sc-60031C$.

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

group X sPLA $_2$ siRNA (m) is recommended for the inhibition of group X sPLA $_2$ expression in mouse 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

group X sPLA₂ (E-4): sc-514324 is recommended as a control antibody for monitoring of group X sPLA₂ 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG κ 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 group X sPLA $_2$ gene expression knockdown using RT-PCR Primer: group X sPLA $_2$ (m)-PR: sc-60031-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.

PROTOCOLS

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