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# group VI iPLA<sub>2</sub> shRNA (m) Lentiviral Particles: sc-43820-V

## BACKGROUND

Phospholipases catalyze the release of fatty acids from phospholipids. One member of the phospholipase family, iPLA<sub>2</sub>, is detected as a membrane-bound protein with multiple smaller isoforms, which result from alternative splicing. Two isoforms, ankyrin- iPLA<sub>2.1</sub> and <sub>2</sub>, lack the catalytic domain and are thought to be involved in the negative regulation of iPLA<sub>2</sub> activity. The SH-iPLA<sub>2</sub> isoform is cytoplasmically localized. Another phospholipase, sPLA<sub>2</sub>, belongs to a family of secretory phospholipases A<sub>2</sub>, which represent an expanding family of related enzymes. sPLA<sub>2</sub> has both membrane bound and secreted forms that are encoded by a single gene. sPLA<sub>2</sub> is involved in the regulation of phospholipid metabolism in biomembranes and in eicosanoid biosynthesis.

## REFERENCES

1. Scott, D.L., White, S.P., Browning, J.L., Rosa, J.J., Gelb, M.H. and Sigler, P.B. 1991. Structures of free and inhibited human secretory phospholipase A<sub>2</sub> from inflammatory exudate. *Science* 254: 1007-1010.
2. Cupillard, L., Koumanov, K., Mattei, M.G., Lazdunski, M. and Lambeau, G. 1997. Cloning, chromosomal mapping, and expression of a novel human secretory phospholipase A<sub>2</sub>. *J. Biol. Chem.* 272: 15745-15752.
3. Kitadokoro, K., Hagishita, S., Sato, T., Ohtani, M. and Miki, K. 1998. Crystal structure of human secretory phospholipase A<sub>2</sub>-IIA complex with the potent indolizine inhibitor 120-1032. *J. Biochem.* 123: 619-623.
4. Ma, Z., Wang, X., Nowatzke, W., Ramanadham, S. and Turk, J. 1999. Human pancreatic islets express mRNA species encoding two distinct catalytically active isoforms of group VI phospholipase A<sub>2</sub> (iPLA<sub>2</sub>) that arise from an exon-skipping mechanism of alternative splicing of the transcript from the iPLA<sub>2</sub> gene on chromosome 22q13.1. *J. Biol. Chem.* 274: 9607-9616.
5. Larsson-Forsell, P.K., Kennedy, B.P. and Claesson, H.E. 1999. The human calcium-independent phospholipase A<sub>2</sub> gene multiple enzymes with distinct properties from a single gene. *Eur. J. Biochem.* 262: 575-585.

## CHROMOSOMAL LOCATION

Genetic locus: Pla2g6 (mouse) mapping to 15 E1.

## PRODUCT

group VI iPLA<sub>2</sub> shRNA (m) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 µl frozen stock containing 1.0 x 10<sup>6</sup> infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see group VI iPLA<sub>2</sub> siRNA (m): sc-43820 and group VI iPLA<sub>2</sub> shRNA Plasmid (m): sc-43820-SH as alternate gene silencing products.

## STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

## APPLICATIONS

group VI iPLA<sub>2</sub> shRNA (m) Lentiviral Particles is recommended for the inhibition of iPLA<sub>2</sub> expression in mouse cells.

## SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 µl frozen viral stock containing 1.0 x 10<sup>6</sup> infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

## GENE EXPRESSION MONITORING

group VI iPLA<sub>2</sub> (D-4): sc-376563 is recommended as a control antibody for monitoring of iPLA<sub>2</sub> 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 iPLA<sub>2</sub> gene expression knockdown using RT-PCR Primer: group VI iPLA<sub>2</sub> (m)-PR: sc-43820-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

## BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

## RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

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