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### SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

[mail@szabo-scandic.com](mailto:mail@szabo-scandic.com)

[www.szabo-scandic.com](http://www.szabo-scandic.com)

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 

# PhLP siRNA (h): sc-45420

## BACKGROUND

Phosducin-like protein (PhLP, PDCL) is an ethanol-responsive modulator of heterotrimeric G proteins. The protein shares extensive amino acid sequence homology with phosducin (Phd), a phosphoprotein expressed in retina and pineal gland. Both PhLP and Phd regulate G protein signaling by binding to the  $\beta\gamma$  subunits of G proteins. PhLP interacts with  $G_{\beta\gamma}$  via a short C-terminal binding site. Additionally, PhLP acts as a substrate for GRK2 phosphorylation at the same C-terminal binding site between residues 195 and 218. PhLPs may participate directly in the regulation of calcium-evoked exocytosis in adrenal medullary chromaffin cells. Glycosylated PhLP regulates opioid receptor function in mouse brain.

## REFERENCES

1. Miles, M.F., Barhite, S., Sganga, M. and Elliott, M. 1993. Phosducin-like protein: an ethanol-responsive potential modulator of guanine nucleotide-binding protein function. *Proc. Natl. Acad. Sci. USA* 90: 10831-10835.
2. Schroder, S., Bluml, K., Dees, C. and Lohse, M.J. 1997. Identification of a C-terminal binding site for G protein  $\beta\gamma$  subunits in phosducin-like protein. *FEBS Lett.* 401: 243-246.
3. Thibault, C., Feng Wang, J., Charnas, R., Mirel, D., Barhite, S. and Miles, M.F. 1999. Cloning and characterization of the rat and human phosducin-like protein genes: structure, expression and chromosomal localization. *Biochim. Biophys. Acta* 1444: 346-354.
4. Ruiz-Gomez, A., Humrich, J., Murga, C., Quitterer, U., Lohse, M.J. and Mayor, F., Jr. 2000. Phosphorylation of phosducin and phosducin-like protein by G protein-coupled receptor kinase 2. *J. Biol. Chem.* 275: 29724-29730.
5. Gensse, M., Vitale, N., Chasserot-Golaz, S. and Bader, M.F. 2000. Regulation of exocytosis in chromaffin cells by phosducin-like protein, a protein interacting with G protein  $\beta\gamma$  subunits. *FEBS Lett.* 480: 184-188.

## CHROMOSOMAL LOCATION

Genetic locus: PDCL (human) mapping to 9q33.2.

## PRODUCT

PhLP 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PhLP shRNA Plasmid (h): sc-45420-SH and PhLP shRNA (h) Lentiviral Particles: sc-45420-V as alternate gene silencing products.

For independent verification of PhLP (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-45420A, sc-45420B and sc-45420C.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) 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  $\mu$ l of the RNase-free water provided. Resuspension of the siRNA duplex in 330  $\mu$ l of RNase-free water makes a 10  $\mu$ M solution in a 10  $\mu$ M Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

## APPLICATIONS

PhLP siRNA (h) is recommended for the inhibition of PhLP 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  $\mu$ M in 66  $\mu$ 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

PhLP (A-8): sc-376918 is recommended as a control antibody for monitoring of PhLP 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> Hard-set Mounting Medium: sc-359850.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PhLP gene expression knockdown using RT-PCR Primer: PhLP (h)-PR: sc-45420-PR (20  $\mu$ 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.