SANTA CRUZ BIOTECHNOLOGY, INC.

PITPα siRNA (m): sc-40856

BACKGROUND

The lipid binding proteins known as phosphatidylinositol transfer proteins (PITP) facilitate the formation of phosphatidylinositol derived second messenger molecules, which are related to the phospholipase C and phosphoinositide 3-kinase pathways. PITP are ubiquitously expressed proteins that transfer phosphatidylinositol (PI) and phosphatidylcholine (PC) between membranes enriched in PI or PC to membranes that are deficient in PI or PC. PITP mobilizes PI from the endoplasmic recticulum and regulates the release of PI from stored vesicles in the Golgi network. In mammalian cells, three smaller forms of soluble PITP are present, designated PITP α, β and retinal degeneration B (rdgB) β. The gene encoding human rdgB β maps to chromosome 11q13, a region that contains several retinopathy loci, which makes the H-rdgB β gene a candidate for several inherited retinal degenerative diseases.

REFERENCES

1. Guo, J. and Yu, F.X. 1997. Cloning and characterization of human homologue of *Drosophila* retinal degeneration B: a candidate gene for degenerative retinal diseases. Dev. Genet. 20: 235-245.
2. Monaco, M.E., Alexander, R.J., Snoek, G.T., Moldover, N.H., Wirtz, K.W. and Walden, P.D. 1998. Evidence that mammalian phosphatidylinositol transfer protein regulates phosphatidylcholine metabolism. Biochem. J. 335: 175-179.
3. Aikawa, Y., Kuraoka, A., Kondo, H., Kawabuchi, M. and Watanabe, T. 1999. Involvement of PITPnm, a mammalian homologue of *Drosophila* rdgB, in phosphoinositide synthesis on Golgi membranes. J. Biol. Chem. 274: 20569-20577.
4. Fullwood, Y., dos Santos, M. and Hsuan, J.J. 1999. Cloning and characterization of a novel human phosphatidylinositol transfer protein, rdgBbeta. J. Biol. Chem. 274: 31553-31558.
5. Cockcroft, S. 1999. Mammalian phosphatidylinositol transfer proteins: emerging roles in signal transduction and vesicular traffic. Chem. Phys. Lipids 98: 23-33.
6. Viscardi, R.M. and Strauss, K.A. 1999. Developmental changes in phosphatidylinositol transfer protein concentration and phospholipid transfer activities in rat type II cells. Exp. Lung Res. 25: 561-576.

CHROMOSOMAL LOCATION

Genetic locus: Pitpna (mouse) mapping to 11 B5.

PRODUCT

PITPα 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PITPα shRNA Plasmid (m): sc-40856-SH and PITPα shRNA (m) Lentiviral Particles: sc-40856-V as alternate gene silencing products.

For independent verification of PITPα (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-40856A, sc-40856B and sc-40856C.

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

PITPα siRNA (m) is recommended for the inhibition of PITPα 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

PITPα (5F12): sc-13569 is recommended as a control antibody for monitoring of PITPα 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 PITPα gene expression knockdown using RT-PCR Primer: PITPα (m)-PR: sc-40856-PR (20 µl). 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.