

GPI-PLD siRNA (h): sc-43811

BACKGROUND

Phosphatidylinositol-glycan-specific phospholipase D (GPI-PLD) is a high-density lipoprotein-associated protein found on chromosome 6p22.3 that specifically hydrolyzes the inositol phosphate linkage in proteins anchored by phosphatidylinositol-glycans (PI-Gs). GPI-PLD is found in serum, liver, cerebrospinal fluid and in milk. The majority of plasma GPI-PLD appears to be specifically associated with a small, discrete, and minor fraction of lipoproteins containing apoA-I and apoA-IV. Serum GPI-PLD activity is reduced over 75% in systemic inflammatory response syndrome and the downregulation of GPI-PLD could play an important role in the control of proinflammatory responses.

REFERENCES

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2. Stieger, S., et al. 1991. Enzymatic properties of phosphatidylinositol-glycan-specific phospholipase C from rat liver and phosphatidylinositol-glycan-specific phospholipase D from rat serum. *Eur. J. Biochem.* 197: 67-73.
3. Hoener, M.C. and Brodbeck, U. 1992. Phosphatidylinositol-glycan-specific phospholipase D is an amphiphilic glycoprotein that in serum is associated with high-density lipoproteins. *Eur. J. Biochem.* 206: 747-757.
4. Scallan, B.J., et al. 1992. A novel strategy for secreting proteins: use of phosphatidylinositol-glycan-specific phospholipase D to release chimeric phosphatidylinositol-glycan anchored proteins. *Biotechnology* 10: 550-556.
5. Deeg, M.A., et al. 2001. Increased expression of GPI-specific phospholipase D in mouse models of type 1 diabetes. *Am. J. Physiol. Endocrinol. Metab.* 281: E147-E154.
6. Deeg, M.A., et al. 2001. GPI-specific phospholipase D associates with an apoA⁻¹ and apoA-IV-containing complex. *J. Lipid Res.* 42: 442-451.
7. Du, X. and Low, M.G. 2001. Downregulation of glycosylphosphatidylinositol-specific phospholipase D induced by lipopolysaccharide and oxidative stress in the murine monocyte-macrophage cell line RAW 264.7. *Infect. Immun.* 69: 3214-3223.

CHROMOSOMAL LOCATION

Genetic locus: GPLD1 (human) mapping to 6p22.3.

PRODUCT

GPI-PLD 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GPI-PLD shRNA Plasmid (h): sc-43811-SH and GPI-PLD shRNA (h) Lentiviral Particles: sc-43811-V as alternate gene silencing products.

For independent verification of GPI-PLD (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-43811A, sc-43811B and sc-43811C.

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

GPI-PLD siRNA (h) is recommended for the inhibition of GPI-PLD 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 μ M in 66 μ 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

GPI-PLD (D-10): sc-365096 is recommended as a control antibody for monitoring of GPI-PLD 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 κ BP-HRP: sc-516102 or m-IgG κ 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-IgG κ BP-FITC: sc-516140 or m-IgG 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 GPI-PLD gene expression knockdown using RT-PCR Primer: GPI-PLD (h)-PR: sc-43811-PR (20 μ l, 485 bp). 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.