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GPI-PLD siRNA (m): sc-41625

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

1. Scallan, B.J., et al. 1991. Primary structure and functional activity of a phosphatidylinositol-glycan-specific phospholipase D. *Science* 252: 446-448.
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: 147-154.

CHROMOSOMAL LOCATION

Genetic locus: *Gpld1* (mouse) mapping to 13 A3.1.

PRODUCT

GPI-PLD 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 GPI-PLD shRNA Plasmid (m): sc-41625-SH and GPI-PLD shRNA (m) Lentiviral Particles: sc-41625-V as alternate gene silencing products.

For independent verification of GPI-PLD (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-41625A, sc-41625B and sc-41625C.

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.

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 (m) is recommended for the inhibition of GPI-PLD 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 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 (m)-PR: sc-41625-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.