

PIG-M siRNA (h): sc-88676

BACKGROUND

Phosphatidylinositol-glycans (PIGs) are multi-pass transmembrane proteins that localize to the endoplasmic reticulum. PIGs exhibit various functions but all are crucial for the biosynthesis of the glycosylphosphatidylinositol (GPI)-anchor. Some PIG proteins are components of the GPI transamidase complex and play a role in the recognition of either the GPI attachment signal or the lipid portion of GPI. Other PIGs belong to the glycosyltransferase complex (GPI-N-acetylglucosaminyltransferase or GPI-GnT) and function in the transfer of N-acetylglucosamine (GlcNAc) to phosphatidylinositol (PI). A variety of other PIGs play distinct roles in GPI synthesis. PIG-M (phosphatidylinositol glycan anchor biosynthesis, class M), also known as GPI-MT-I, is a 423 amino acid mannosyltransferase involved in glycosylphosphatidylinositol-anchor biosynthesis. Mutations in the gene encoding PIG-M leads to glycosylphosphatidylinositol deficiency (GPID), an autosomal recessive trait that results in a propensity to venous thrombosis and seizures.

REFERENCES

- Inoue, N., et al. 1996. PIG-C, one of the three human genes involved in the first step of glycosylphosphatidylinositol biosynthesis is a homologue of *Saccharomyces cerevisiae* GPI2. *Biochem. Biophys. Res. Commun.* 226: 193-199.
- Watanabe, R., et al. 1998. The first step of glycosylphosphatidylinositol biosynthesis is mediated by a complex of PIG-A, PIG-H, PIG-C and GPI1. *EMBO J.* 17: 877-885.
- Watanabe, R., et al. 2000. Initial enzyme for glycosylphosphatidylinositol biosynthesis requires PIG-P and is regulated by DPM2. *EMBO J.* 19: 4402-4411.
- Tiede, A., et al. 2000. Characterisation of the enzymatic complex for the first step in glycosylphosphatidylinositol biosynthesis. *Int. J. Biochem. Cell Biol.* 32: 339-350.
- Maeda, Y., et al. 2001. PIG-M transfers the first mannose to glycosylphosphatidylinositol on the luminal side of the ER. *EMBO J.* 20: 250-261.

CHROMOSOMAL LOCATION

Genetic locus: PIGM (human) mapping to 1q23.2.

PRODUCT

PIG-M 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 PIG-M shRNA Plasmid (h): sc-88676-SH and PIG-M shRNA (h) Lentiviral Particles: sc-88676-V as alternate gene silencing products.

For independent verification of PIG-M (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-88676A, sc-88676B and sc-88676C.

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

PIG-M siRNA (h) is recommended for the inhibition of PIG-M 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PIG-M gene expression knockdown using RT-PCR Primer: PIG-M (h)-PR: sc-88676-PR (20 μ 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.