

# PIG-Y siRNA (m): sc-76138

## 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-Y is a component of the GPI-GnT complex which is responsible for the first step in GPI synthesis, the transfer of GlcNAc to PI from UDP-GlcNAc. PIG-Y specifically interacts with PIG-A and this interaction is critical for the activity of GPI-GnT. The loss of PIG-Y severely affects the biosynthesis of the GPI-anchor.

## REFERENCES

- Hong, Y., Ohishi, K., Watanabe, R., Endo, Y., Maeda, Y. and Kinoshita, T. 1999. GPI1 stabilizes an enzyme essential in the first step of glycosylphosphatidylinositol biosynthesis. *J. Biol. Chem.* 274: 18582-18588.
- Tiede, A., Nischan, C., Schubert, J. and Schmidt, R.E. 2000. Characterisation of the enzymatic complex for the first step in glycosylphosphatidylinositol biosynthesis. *Int. J. Biochem. Cell Biol.* 32: 339-350.
- Ikezawa, H. 2002. Glycosylphosphatidylinositol (GPI)-anchored proteins. *Biol. Pharm. Bull.* 25: 409-417.
- Murakami, Y., Siripanyaphinyo, U., Hong, Y., Tashima, Y., Maeda, Y. and Kinoshita, T. 2005. The initial enzyme for glycosylphosphatidylinositol biosynthesis requires PIG-Y, a seventh component. *Mol. Biol. Cell* 16: 5236-5246.
- Newman, H.A., Romeo, M.J., Lewis, S.E., Yan, B.C., Orlean, P. and Levin, D.E. 2005. Gpi19, the *Saccharomyces cerevisiae* homologue of mammalian PIG-P, is a subunit of the initial enzyme for glycosylphosphatidylinositol anchor biosynthesis. *Eukaryotic Cell* 4: 1801-1807.
- Brodsky, R.A. 2006. New insights into paroxysmal nocturnal hemoglobinuria. *Hematology Am. Soc. Hematol. Educ. Program* 2006: 24-28.
- Orlean, P. and Menon, A.K. 2007. Thematic review series: lipid posttranslational modifications. GPI anchoring of protein in yeast and mammalian cells, or: how we learned to stop worrying and love glycopospholipids. *J. Lipid Res.* 48: 993-1011.
- Li, H., Zhou, H., Luo, Y., Ouyang, H., Hu, H. and Jin, C. 2007. Glycosylphosphatidylinositol (GPI) anchor is required in *Aspergillus fumigatus* for morphogenesis and virulence. *Mol. Microbiol.* 64: 1014-1027.
- Pittet, M. and Conzelmann, A. 2007. Biosynthesis and function of GPI proteins in the yeast *Saccharomyces cerevisiae*. *Biochim. Biophys. Acta* 1771: 405-420.

## PROTOCOLS

See our web site at [www.scbt.com](http://www.scbt.com) for detailed protocols and support products.

## CHROMOSOMAL LOCATION

Genetic locus: Pigy (mouse) mapping to 6 B3.

## PRODUCT

PIG-Y 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PIG-Y shRNA Plasmid (m): sc-76138-SH and PIG-Y shRNA (m) Lentiviral Particles: sc-76138-V as alternate gene silencing products.

For independent verification of PIG-Y (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-76138A, sc-76138B and sc-76138C.

## 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

PIG-Y siRNA (m) is recommended for the inhibition of PIG-Y 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  $\mu$ 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.

## RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PIG-Y gene expression knockdown using RT-PCR Primer: PIG-Y (m)-PR: sc-76138-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.