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PIG-N siRNA (m): sc-152254

BACKGROUND

Several cell surface proteins are attached to the membrane through their C-terminal domain and a glycosylphosphatidylinositol (GPI) moiety. Phosphatidylinositol-glycans (PIGs) are multi-pass transmembrane proteins that localize to the endoplasmic reticulum. PIGs are crucial for the synthesis of N-acetylglucosaminyl-phosphatidylinositol, a very early intermediate in GPI-anchor biosynthesis. PIGs play a role in the recognition of either the GPI attachment signal or the lipid portion of GPI. PIG-N (phosphatidylinositol-glycan biosynthesis class N protein), also known as GPI ethanolamine phosphate transferase 1 and MCD4 homolog, is a 931 amino acid enzyme of the endoplasmic reticulum that transfers ethanolamine phosphate to the first α -1,4-linked mannose of the glycosylphosphatidylinositol precursor of GPI-anchor. The gene encoding PIG-N is localized near a region of human chromosome 18 that may be implicated in chronic recurrent multifocal osteomyelitis.

REFERENCES

1. Yeh, E.T., et al. 1994. Biosynthesis and processing of the glycosylphosphatidylinositol anchor in mammalian cells. *Semin. Immunol.* 6: 73-80.
2. Hong, Y., et al. 1999. PIG-N, a mammalian homologue of yeast Mcd4p, is involved in transferring phosphoethanolamine to the first mannose of the glycosylphosphatidylinositol. *J. Biol. Chem.* 274: 35099-35106.
3. Gaynor, E.C., et al. 1999. MCD4 encodes a conserved endoplasmic reticulum membrane protein essential for glycosylphosphatidylinositol anchor synthesis in yeast. *Mol. Biol. Cell* 10: 627-648.
4. Barz, W.P. and Walter, P. 1999. Two endoplasmic reticulum (ER) membrane proteins that facilitate ER-to-Golgi transport of glycosylphosphatidylinositol-anchored proteins. *Mol. Biol. Cell* 10: 1043-1059.
5. Yada, T., et al. 2001. Its8, a fission yeast homolog of Mcd4 and PIG-N, is involved in GPI anchor synthesis and shares an essential function with calcineurin in cytokinesis. *J. Biol. Chem.* 276: 13579-13586.

CHROMOSOMAL LOCATION

Genetic locus: Pign (mouse) mapping to 1 E2.1.

PRODUCT

PIG-N 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 PIG-N shRNA Plasmid (m): sc-152254-SH and PIG-N shRNA (m) Lentiviral Particles: sc-152254-V as alternate gene silencing products.

For independent verification of PIG-N (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-152254A, sc-152254B and sc-152254C.

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-N siRNA (m) is recommended for the inhibition of PIG-N 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.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PIG-N gene expression knockdown using RT-PCR Primer: PIG-N (m)-PR: sc-152254-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.