SANTA CRUZ BIOTECHNOLOGY, INC.

PIG-U siRNA (h): sc-61351

BACKGROUND

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 glycosylphosphatidylinositol (GPI)-anchor biosynthesis. PIGs play a role in the recognition of either the GPI attachment signal or the lipid portion of GPI. PIG-U (CDC91L1) is a critical part of GPI transamidase (GPIT), a multisubunit membrane-bound complex also consisting of Gaa1, Gpi8, PIG-S, and PIG-T. GPIT recognizes C-terminal signal sequences on proproteins and replaces them with specific GPI lipids. The PIG-U gene is oncogenic and is implicated in the development of human bladder cancer. Overexpression of PIG-U causes increased expression of the urokinase receptor (uPAR), a GPI-anchored protein, thereby amplifying signal transducer and activator of transcription (STAT-3) phosphorylation in bladder cancer cells, which may lead to cancer.

REFERENCES

1. Lewis, A.J., et al. 1979. The anti-inflammatory profile of dapsone in animal models of inflammation. Agents Actions 8: 578-586.
2. Rondeau, E., et al. 1991. Nordihydroguaiaretic acid inhibits urokinase synthesis by phorbol myristate acetate-stimulated LLC-PK1 cells. Biochim. Biophys. Acta 1055: 165-172.
3. Hong, Y., et al. 2003. Human PIG-U and yeast Cdc91p are the fifth subunit of GPI transamidase that attaches GPI-anchors to proteins. Mol. Biol. Cell 14: 1780-1789.
4. Nagamune, K., et al. 2003. GPI transamidase of *Trypanosoma brucei* has two previously uncharacterized (trypanosomatid transamidase 1 and 2) and three common subunits. Proc. Natl. Acad. Sci. USA 100: 10682-10687.
5. Guo, Z., et al. 2004. CDC91L1 (PIG-U) is a newly discovered oncogene in human bladder cancer. Nat. Med. 10: 374-381.
6. Vainauskas, S. and Menon, A.K. 2004. A conserved proline in the last transmembrane segment of Gaa1 is required for glycosylphosphatidylinositol (GPI) recognition by GPI transamidase. J. Biol. Chem. 279: 6540-6545.
7. Montie, J.E., et al. 2005. CDC91L1 (PIG-U) is a newly discovered oncogene in human bladder cancer. J. Urol. 174: 869-870.

CHROMOSOMAL LOCATION

Genetic locus: PIGU (human) mapping to 20q11.22.

PRODUCT

PIG-U 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PIG-U shRNA Plasmid (h): sc-61351-SH and PIG-U shRNA (h) Lentiviral Particles: sc-61351-V as alternate gene silencing products.

For independent verification of PIG-U (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-61351A, sc-61351B and sc-61351C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PIG-U siRNA (h) is recommended for the inhibition of PIG-U 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 µM in 66 µl. 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-U gene expression knockdown using RT-PCR Primer: PIG-U (h)-PR: sc-61351-PR (20 µl, 589 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.