

GPAA1 siRNA (m): sc-60716

BACKGROUND

Glycosylphosphatidylinositol (GPI) acts as a membrane anchor for cell surface proteins. Glycosylphosphatidylinositol anchor attachment 1 protein (GPAA1), also designated GPI anchor attachment protein 1 or GAA1 protein homolog, is a membrane protein localized to the endoplasmic reticulum which is involved in GPI-anchor biosynthesis. GPAA1 is crucial for GPI-anchoring of precursor proteins and catalyzes the attachment of GPI to proteins containing a C-terminal GPR attachment signal. GAA1 contains an N-terminal signal sequence, one cAMP- and cGMP-dependent protein kinase phosphorylation site, two potential N-glycosylation sites, one leucine zipper pattern and eight putative transmembrane domains. GPAA1 is ubiquitously expressed and shows higher levels of expression in fetal tissues than in adult tissues.

REFERENCES

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2. Online Mendelian Inheritance in Man, OMIM[™]. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 603048. World Wide Web URL: <http://www.ncbi.nlm.nih.gov/omim/>
3. Azzouz, N., et al. 2005. Removal of phospholipid contaminants through precipitation of glycosylphosphatidylinositols. *Anal. Biochem.* 343: 152-158.
4. Hilvo, M., et al. 2005. Characterization of CA XV, a new GPI-anchored form of carbonic anhydrase. *Biochem. J.* 392: 83-92.
5. Hutchinson, T.E., et al. 2005. Phospholipase C sensitive GPI-anchored proteins of goat sperm: possible role in sperm protection. *Anim. Reprod. Sci.* 88: 271-286.
6. Joh, T., et al. 2005. *Helicobacter pylori* eradication decreases the expression of glycosylphosphatidylinositol-anchored complement regulators, decay-accelerating factor and homologous restriction factor 20, in human gastric epithelium. *J. Gastroenterol. Hepatol.* 20: 1344-1351.
7. Wei, Z., et al. 2005. *Listeria monocytogenes* phosphatidylinositol-specific phospholipase C has evolved for virulence by greatly reduced activity on GPI anchors. *Proc. Natl. Acad. Sci. USA* 102: 12927-12931.

CHROMOSOMAL LOCATION

Genetic locus: Gpaa1 (mouse) mapping to 15 D3.

PRODUCT

GPAA1 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 GPAA1 shRNA Plasmid (m): sc-60716-SH and GPAA1 shRNA (m) Lentiviral Particles: sc-60716-V as alternate gene silencing products.

For independent verification of GPAA1 (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-60716A, sc-60716B and sc-60716C.

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

GPAA1 siRNA (m) is recommended for the inhibition of GPAA1 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

GPAA1 (F-12): sc-373710 is recommended as a control antibody for monitoring of GPAA1 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 GPAA1 gene expression knockdown using RT-PCR Primer: GPAA1 (m)-PR: sc-60716-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.

PROTOCOLS

See our web site at www.scbt.com for detailed protocols and support products.