SANTA CRUZ BIOTECHNOLOGY, INC.

GPA2 siRNA (m): sc-60714

BACKGROUND

Glycoprotein hormone α2 subunit (GPA2) belongs to the dimeric glycoprotein hormones α chain family. GPA2 is an N-linked glycosylated secreted protein with ten cysteine residues likely involved in cysteine-knot formation. It forms a heterodimer with glycoprotein hormone β5 subunit (GPB5), also called thyrostimulin hormone, and activates thyroid stimulating hormone receptor (also designated thyrotropin receptor or TSHR), which increases cAMP production and stimulates the thymus. GPA2 and GPB5 are both evolutionarily conserved and GPA2 may serve as a scaffold for GPB5 for downstream G protein-coupled signaling. GPA2 demonstrates ubiquitious expression and co-localizes with GPB5 in the eye, testis and pituitary (GPA2 detected in the anterior lobe).

REFERENCES

1. Hsu, S.Y., et al. 2002. Evolution of glycoprotein hormone subunit genes in bilateral metazoa: identification of two novel human glycoprotein hormone subunit family genes, GPA2 and GPB5. Mol. Endocrinol. 16: 1538-1551.
2. Maidan, M.M., et al. 2005. The G protein-coupled receptor Gpr1 and the G_α protein GPA2 act through the cAMP-protein kinase A pathway to induce morphogenesis in *Candida albicans*. Mol. Biol. Cell 16: 1971-1986.
3. Harashima, T., et al. 2005. G_α subunit GPA2 recruits kelch repeat subunits that inhibit receptor-G protein coupling during cAMP-induced dimorphic transitions in *Saccharomyces cerevisiae*. Mol. Biol. Cell 16: 4557-4571.
4. Ivey, F.D., et al. 2005. Direct activation of fission yeast adenylate cyclase by the GPA2 G_α of the glucose signaling pathway. Proc. Natl. Acad. Sci. USA 102: 6108-6113.
5. Sudo, S., et al. 2005. Heterodimeric fly glycoprotein hormone-α2 (GPA2) and glycoprotein hormone-β5 (GPB5) activate fly leucine-rich repeat-containing G protein-coupled receptor-1 (DLGR1) and stimulation of human thyrotropin. Endocrinology 146: 3596-3604.
6. Nagasaki, H., et al. 2006. Differential expression of the thyrostimulin subunits, glycoprotein α2 and β5 in the rat pituitary. J. Mol. Endocrinol. 37: 39-50.

CHROMOSOMAL LOCATION

Genetic locus: Gpha2 (mouse) mapping to 19 A.

PRODUCT

GPA2 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GPA2 shRNA Plasmid (m): sc-60714-SH and GPA2 shRNA (m) Lentiviral Particles: sc-60714-V as alternate gene silencing products.

For independent verification of GPA2 (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-60714A, sc-60714B and sc-60714C.

RESEARCH USE

For research use only, not for use in diagnostic procedures.

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

GPA2 siRNA (m) is recommended for the inhibition of GPA2 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 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.

GENE EXPRESSION MONITORING

GPA2 (G-3): sc-390194 is recommended as a control antibody for monitoring of GPA2 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 GPA2 gene expression knockdown using RT-PCR Primer: GPA2 (m)-PR: sc-60714-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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