

# GPA2 siRNA (h): sc-60713

## BACKGROUND

Glycoprotein hormone  $\alpha 2$  subunit (GPA2) belongs to the dimeric glycoprotein hormones  $\alpha$  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  $\beta 5$  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 ubiquitous expression and co-localizes with GPB5 in the eye, testis and pituitary (GPA2 detected in the anterior lobe).

## REFERENCES

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2. Maidan, M.M., et al. 2005. The G protein-coupled receptor Gpr1 and the  $G_{\alpha}$  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.  $G_{\alpha}$  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  $G_{\alpha}$  of the glucose signaling pathway. *Proc. Natl. Acad. Sci. USA* 102: 6108-6113.
5. Sudo, S., et al. 2005. Heterodimeric fly glycoprotein hormone- $\alpha 2$  (GPA2) and glycoprotein hormone- $\beta 5$  (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  $\alpha 2$  and  $\beta 5$  in the rat pituitary. *J. Mol. Endocrinol.* 37: 39-50.

## CHROMOSOMAL LOCATION

Genetic locus: GPA2 (human) mapping to 11q13.1.

## PRODUCT

GPA2 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  $\mu$ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GPA2 shRNA Plasmid (h): sc-60713-SH and GPA2 shRNA (h) Lentiviral Particles: sc-60713-V as alternate gene silencing products.

For independent verification of GPA2 (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-60713A, sc-60713B and sc-60713C.

## 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  $\mu$ 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

GPA2 siRNA (h) is recommended for the inhibition of GPA2 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  $\mu$ 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.

## 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-IgG $\kappa$  BP-HRP: sc-516102 or m-IgG $\kappa$  BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker<sup>™</sup> Molecular Weight Standards: sc-2035, UltraCruz<sup>®</sup> Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-IgG $\kappa$  BP-FITC: sc-516140 or m-IgG $\kappa$  BP-PE: sc-516141 (dilution range: 1:50-1:200) with UltraCruz<sup>®</sup> Mounting Medium: sc-24941 or UltraCruz<sup>®</sup> 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 (h)-PR: sc-60713-PR (20  $\mu$ l). 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](http://www.scbt.com) for detailed protocols and support products.