GnRH I siRNA (h): sc-39542



The Power to Ouestion

BACKGROUND

Human reproduction is controlled by the hypothalaic-pituitary gonadal axis laid down early in fetal development. Gonadotropin releasing hormone (GnRH), also known as GnRH-associated peptide, luteinizing hormone releasing hormone (LHRH), luliberin or gonadorelin, is a decapeptide that is an important molecule in the hypothalamic-pituitary-gonadal axis control circuit. GnRH is produced by hypothalamic neurons and secreted in a pulsatile manner into the capillary plexus of the medianeminence. GnRH affects the release of lutenizing hormone and follicle stimulating hormone from gonadotropic cells in the anterior pituitary. In addition to hypothalamic GnRH (GnRH I), a second GnRH form (GnRH II) functions primarily in the midbrain. GnRH is expressed in the acrosomal region of human sperm and in the anterior pituitary tissue and cancer cells. Unlike GnRH I, GnRH II is highly expressed outside the brain, particularly in the kidney, bone marrow and prostate, suggesting that it may have multiple functions. GnRH binds to a specific G protein-coupled receptor in the pituitary to regulate synthesis and secretion of gonadotropins.

REFERENCES

- 1. Seeburg, P.H., et al. 1984. Characterization of cDNA for precursor of human luteinizing hormone releasing hormone. Nature 311: 666-668.
- Grosse, R., et al. 1997. Inhibition of gonadotropin-releasing hormone receptor signaling by expression of a splice variant of the human receptor. Mol. Endocrinol. 11: 1305-1318.
- 3. White, R.B., et al. 1998. Second gene for gonadotropin-releasing hormone in humans. Proc. Natl. Acad. Sci. USA 95: 305-309.
- 4. Goto, T., et al. 1999. Gonadotropin-releasing hormone agonist has the ability to induce increased matrix metalloproteinase (MMP)-2 and membrane type 1-MMP expression in corpora lutea, and structural luteolysis in rats. J. Endocrinol. 161: 393-402.
- Lee, C.Y., et al. 2000. Immunoidentification of gonadotropin releasing hormone receptor in human sperm, pituitary and cancer cells. Am. J. Reprod. Immunol. 44: 170-177.
- 6. Maudsley, S., et al. 2004. Gonadotropin-releasing hormone (GnRH) antagonists promote proapoptotic signaling in peripheral reproductive tumor cells by activating a $G_{\alpha i}$ -coupling state of the type I GnRH receptor. Cancer Res. 64: 7533-7544.

CHROMOSOMAL LOCATION

Genetic locus: GNRH1 (human) mapping to 8p21.2.

PRODUCT

GnRH I 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see GnRH I shRNA Plasmid (h): sc-39542-SH and GnRH I shRNA (h) Lentiviral Particles: sc-39542-V as alternate gene silencing products.

For independent verification of GnRH I (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-39542A, sc-39542B and sc-39542C.

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

GnRH I siRNA (h) is recommended for the inhibition of GnRH I 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.

GENE EXPRESSION MONITORING

GnRH I (A-4): sc-271918 is recommended as a control antibody for monitoring of GnRH I 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 GnRH I gene expression knockdown using RT-PCR Primer: GnRH I (h)-PR: sc-39542-PR (20 μ I, 479 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.

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