



TSHR siRNA (h): sc-36754

BACKGROUND

Various hormones are secreted from the anterior pituitary during development and growth, including thyroid-stimulating hormone (TSH, also known as thyrotropin), follicle-stimulating hormone (FSH) and leutinizing hormone (LH). TSH, FSH, and LH are heterodimers formed from a common α chain and a unique β chain. TSH is a glycoprotein involved in the control of thyroid structure and metabolism, which stimulates the release of the thyroid hormones. TSH is regulated by thyroid hormone (T3) and various retinoid compounds. TSH binds to the thyroid-stimulating hormone receptor (TSHR), which is cleaved into two subunits, A and B, and plays a major role in regulating thyroid function. The third cytoplasmic loop of TSHR has been identified as critical for its role in regulating inositol phosphate and cAMP formation. In Graves disease, an autoimmune disorder, TSHR is activated by autoantibodies, which may be stimulated by the cleavage of the A and B subunits.

REFERENCES

1. Kosugi, S., et al. 1993. Substitutions of different regions of the third cytoplasmic loop of the thyrotropin (TSH) receptor have selective effects on constitutive, TSH, and TSH receptor autoantibody-stimulated phosphoinositide and 3',5'-cyclic adenosine monophosphate signal generation. *Mol. Endocrinol.* 7: 1009-1020.
2. Breen, J.J., et al. 1997. The rat TSH β gene contains distinct response elements for regulation by retinoids and thyroid hormone. *Mol. Cell. Endocrinol.* 131: 137-146.
3. Sanders, J., et al. 1997. Understanding the thyrotropin receptor function-structure relationship. *Baillieres Clin. Endocrinol. Metab.* 11: 451-479.

CHROMOSOMAL LOCATION

Genetic locus: TSHR (human) mapping to 14q31.1.

PRODUCT

TSHR 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 TSHR shRNA Plasmid (h): sc-36754-SH and TSHR shRNA (h) Lentiviral Particles: sc-36754-V as alternate gene silencing products.

For independent verification of TSHR (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-36754A, sc-36754B and sc-36754C.

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

TSHR siRNA (h) is recommended for the inhibition of TSHR 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 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

TSHR (C-10): sc-515556 is recommended as a control antibody for monitoring of TSHR 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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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 TSHR gene expression knockdown using RT-PCR Primer: TSHR (h)-PR: sc-36754-PR (20 μ l, 485 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Kim, S., et al. 2018. Di-2-ethylhexylphthalate promotes thyroid cell proliferation and DNA damage through activating thyrotropin-receptor-mediated pathways *in vitro* and *in vivo*. *Food Chem. Toxicol.* 124: 265-272.
2. Yu, Z., et al. 2022. Follicle stimulating hormone promotes production of renin through its receptor in juxtaglomerular cells of kidney. *Diabetol. Metab. Syndr.* 14: 65.

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.