

TR α siRNA (h): sc-36707

BACKGROUND

Thyroid hormone nuclear receptors (TRs) are ligand-dependent transcription factors which regulate and control many metabolic and developmental processes. There are two genes encoding TRs identified to date, TR α and TR β . TRs bind to thyroid hormone response elements (TREs) with half-site binding motifs in the orientation of palindromes, direct repeats or inverted palindromes. The affinities of binding are both variable and influenced differentially by 3,5,3'-triiodo-L-thyronine (T3). Transcriptional regulation by TRs is also modulated by heterodimerization with TR nuclear accessory proteins, the most extensively characterized of which are the retinoid X receptors (RXR α , RXR β and RXR γ). The TR α isoform TR α 1 can display both a nuclear and undefined cytoplasmic location, and is the only TR that is imported into the mitochondrial matrix. TR α 2 is a C-terminal variant of TR α 1 that does not bind thyroid hormones (THs) and weakly binds DNA. TR α 2 acts as a dominant negative antagonist of TH signalling.

REFERENCES

1. Näär, A., et al. 1991. The orientation and spacing of core DNA-binding motifs dictate selective transcriptional responses to three nuclear receptors. *Cell* 65: 1267-1271.
2. Lazar, M.A. 1993. Thyroid hormone receptors: multiple forms, multiple possibilities. *Endocrinol. Rev.* 14: 184-193.
3. Meier, C.A., et al. 1993. Interaction of human TR β 1 and its mutants with DNA and RXR β . T3 response element-dependent dominant negative potency. *J. Clin. Invest.* 92: 1986-1993.
4. Zhang, X.K., et al. 1993. Hetero- and homodimeric receptors in thyroid hormone and vitamin A action. *Receptor* 3: 183-191.
5. Bhat, M.K., et al. 1994. Phosphorylation enhances the target gene sequence-dependent dimerization of thyroid hormone receptor with retinoid X receptor. *Proc. Natl. Acad. Sci. USA* 91: 7927-7931.
6. Sugawara, A., et al. 1994. Phosphorylation selectively increases triiodothyronine receptor homodimer binding to DNA. *J. Biol. Chem.* 269: 433-437.
7. Mangelsdorf, D.J., et al. 1994. The retinoid receptors. In Sporn, M.B., et al, eds. *The Retinoids: Biology, Chemistry, and Medicine*. New York: Raven Press, Ltd., 319-349.

CHROMOSOMAL LOCATION

Genetic locus: THRA (human) mapping to 17q21.1.

PRODUCT

TR α 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 TR α shRNA Plasmid (h): sc-36707-SH and TR α shRNA (h) Lentiviral Particles: sc-36707-V as alternate gene silencing products.

For independent verification of TR α (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-36707A, sc-36707B and sc-36707C.

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

TR α siRNA (h) is recommended for the inhibition of TR α 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

TR α 1/ β 1 (C4): sc-740 is recommended as a control antibody for monitoring of TR α 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 MarkerTM 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 TR α gene expression knockdown using RT-PCR Primer: TR α (h)-PR: sc-36707-PR (20 μ l, 419 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Flamini, M.I., et al. 2017. Thyroid hormone controls breast cancer cell movement via integrin $\alpha_v\beta_3$ /SRC/FAK/PI3-kinases. *Horm. Cancer* 8: 16-27.

RESEARCH USE

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