RORγ siRNA (h): sc-38880



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BACKGROUND

The nuclear orphan receptors $ROR\alpha$ and $ROR\gamma$ are members of the nuclear hormone receptor superfamily. This family acts by directly associating with DNA sequences known as hormone response elements (HREs) and typically bind DNA as either homo- or heterodimers. $ROR\alpha$ and $ROR\gamma$ are unique in that they bind DNA as monomers. $ROR\alpha$ has multiple isoforms that share common DNA and putative ligand-binding domains, but differ in their amino terminal domains, which are generated by alternative RNA processing. $ROR\gamma$ comprises a 560 amino acid protein that shares 50% amino acid identity with $ROR\alpha$ and is most highly expressed in skeletal muscle. Although these proteins are considered "orphan receptors", due to a lack of defined ligands, experimental evidence has shown that melatonin may be the natural ligand for these nuclear receptors. The gene encoding $ROR\alpha$ maps to chromosome 15q22.2 and the gene encoding $ROR\gamma$ maps to chromosome 1q21.3.

REFERENCES

- Giguere, V., et al. 1994. Isoform-specific amino-terminal domains dictate DNA-binding properties of RORα, a novel family of orphan hormone nuclear receptors. Genes Dev. 8: 538-543.
- Hirose, T., et al. 1994. RORγ: the third member of ROR/RZR orphan receptor subfamily that is highly expressed in skeletal muscle. Biochem. Biophys. Res. Commun. 205: 1976-1983.
- 3. Carlberg, C., et al. 1995. The orphan receptor family RZR/ROR, melatonin and 5-lipoxygenase: an unexpected relationship. J. Pineal Res. 18: 171-178.
- Mangelsdorf, D.J., et al. 1995. The nuclear receptor superfamily: the second decade. Cell 83: 835-839.

CHROMOSOMAL LOCATION

Genetic locus: RORC (human) mapping to 1q21.3.

PRODUCT

ROR γ 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 ROR γ shRNA Plasmid (h): sc-38880-SH and ROR γ shRNA (h) Lentiviral Particles: sc-38880-V as alternate gene silencing products.

For independent verification of ROR γ (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-38880A, sc-38880B and sc-38880C.

STORAGE AND RESUSPENSION

Store lyophilized siRNA duplex at -20 $^{\circ}$ C with desiccant. Stable for at least one year from the date of shipment. Once resuspended, store at -20 $^{\circ}$ 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

 $ROR\gamma$ siRNA (h) is recommended for the inhibition of $ROR\gamma$ 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

ROR γ (D-4): sc-365476 is recommended as a control antibody for monitoring of ROR γ 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 ROR γ gene expression knockdown using RT-PCR Primer: ROR γ (h)-PR: sc-38880-PR (20 μ I, 428 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

- 1. Ou, Z., et al. 2013. Regulation of the human hydroxysteroid sulfotransferase (SULT2A1) by ROR α and ROR γ and its potential relevance to human liver diseases. Mol. Endocrinol. 27: 106-115.
- Chaudhari, S.N., et al. 2021. A microbial metabolite remodels the gut-liver axis following bariatric surgery. Cell Host Microbe 29: 408-424.e7.

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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