

PPAR α siRNA (h2): sc-44323

BACKGROUND

Peroxisome proliferator-activated receptors (PPARs) are nuclear hormone receptors that can be activated by a variety of compounds including fibrates, thiazolidinediones, prostaglandins and fatty acids. Three PPAR subtypes, designated PPAR α , PPAR β (also designated PPAR δ) and PPAR γ , have been described. PPARs promote transcription by forming heterodimers with members of the retinoid X receptor (RXR) family of steroid receptors and binding to specific DNA motifs termed PPAR-response elements (PPREs). PPAR α is abundant in primary hepatocytes where it regulates the expression of proteins involved in fatty acid metabolism. PPAR β is the most widely distributed subtype and is often expressed at high levels. PPAR γ is predominantly seen in adipose tissue where it plays a critical role in regulating adipocyte differentiation. Interestingly, both the orphan nuclear hormone receptor LXR α and thyroid receptor (TR) have been shown to act as antagonists of PPAR α /RXR α binding to PPREs.

REFERENCES

1. Brun, R.P., et al. 1996. Differential activation of adipogenesis by multiple PPAR isoforms. *Genes Dev.* 10: 974-984.
2. Mansen, A., et al. 1996. Expression of the peroxisome proliferator-activated receptor (PPAR) in the mouse colonic mucosa. *Biochem. Biophys. Res. Commun.* 222: 844-851.
3. Lemberger, T., et al. 1996. Expression of the peroxisome proliferator-activated receptor α gene is stimulated by stress and follows a diurnal rhythm. *J. Biol. Chem.* 271: 1764-1769.
4. Braissant, O., et al. 1996. Differential expression of peroxisome proliferator-activated receptors (PPARs): tissue distribution of PPAR α , β , and γ in the adult rat. *Endocrinology* 137: 354-366.
5. Sterchele, P.F., et al. 1996. Regulation of peroxisome proliferator-activated receptor- α mRNA in rat liver. *Arch. Biochem. Biophys.* 326: 281-289.
6. Miyata, K.S., et al. 1996. The orphan nuclear hormone receptor LXR α interacts with the peroxisome proliferator-activated receptor and inhibits peroxisome proliferator signaling. *J. Biol. Chem.* 271: 9189-9192.

CHROMOSOMAL LOCATION

Genetic locus: PPARA (human) mapping to 22q13.31.

PRODUCT

PPAR α siRNA (h2) 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 PPAR α shRNA Plasmid (h2): sc-44323-SH and PPAR α shRNA (h2) Lentiviral Particles: sc-44323-V as alternate gene silencing products.

For independent verification of PPAR α (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-44323A, sc-44323B and sc-44323C.

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

PPAR α siRNA (h2) is recommended for the inhibition of PPAR 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

PPAR α (H-2): sc-398394 is recommended as a control antibody for monitoring of PPAR 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 Marker[™] 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 PPAR α gene expression knockdown using RT-PCR Primer: PPAR α (h2)-PR: sc-44323-PR (20 μ l). Annealing temperature for the primers should be 55-60 $^{\circ}$ C and the extension temperature should be 68-72 $^{\circ}$ 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.