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SANTA CRUZ BIOTECHNOLOGY, INC.

PPARα siRNA (h2): sc-44323

BACKGROUND

Peroxisome proliferator-activated receptors (PPARs) are nuclear hormone receptors that can be activated by a variety of compounds including fibratus, thiazolidinediones, prostaglandins and fatty acids. Three PPAR subtypes, designated PPARα, PPARβ (also designated PPARδ) and PPARγ, 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). PPARα is abundant in primary hepatocytes where it regulates the expression of proteins involved in fatty acid metabolism. PPARβ is the most widely distributed subtype and is often expressed at high levels. PPARγ is predominantly seen in adipose tissue where it plays a critical role in regulating adipocyte differentiation. Interestingly, both the orphan nuclear hormone receptor LXRα and thyroid receptor (TR) have been shown to act as antagonists of PPARα/RXRα 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 PPARα, -β, and -γ in the adult rat. Endocrinology 137: 354-366.

CHROMOSOMAL LOCATION

Genetic locus: PPARa (human) mapping to 17p12.31.

PRODUCT

PPARα 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PPARα shRNA Plasmid (h2): sc-44323-SH and PPARα shRNA (h2) Lentiviral Particles: sc-44323-V as alternate gene silencing products.

For independent verification of PPARα (h2) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3 nmol of lyophilized siRNA. These include: sc-44323A, sc-44323B and sc-44323C.

RESEARCH USE

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

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

PPARα siRNA (h2) is recommended for the inhibition of PPARα 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

PPARα (H-2): sc-398394 is recommended as a control antibody for monitoring of PPARα 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 (secondary) reagents are recommended: 1) Western Blotting: use goat anti-mouse IgG-HRP: sc-2005 (dilution range: 1:2000-1:32,000) or Cruz Marker™ compatible goat anti-mouse IgG-HRP: sc-2031 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use goat anti-mouse IgG-FITC: sc-2010 (dilution range: 1:100-1:400) or goat anti-mouse IgG-TR: sc-2781 (dilution range: 1:100-1:400) with UltraCruz™ Mounting Medium: sc-24941.

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor PPARα gene expression knockdown using RT-PCR Primer: PPARα (h2)-PR: sc-44323-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.