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SANTA CRUZ BIOTECHNOLOGY, INC.

CYP11B1 siRNA (h): sc-44795

BACKGROUND

The steroid 11β-hydroxylase gene, also designated CYP11B1, is a marker for the functional differentiation of cells in the zonae fasciculata reticularis. The deduced protein CYP11B1 consists of 466 amino acids containing a secretory signal, epidermal growth factor-like repeats, and a proteolytically inactive cathepsin B-related sequence. A related protein, human aldosterone synthase (CYP11B2), is involved in substrate recognition and conversion, with a functionally significant residue 112 in the N-terminal region of human CYP11B2. The inherited disorder glucocorticoid-remediable aldosteronism iscaused by a chimeric gene duplication between the CYP11B1 and CYP11B2 genes. This disorder is characterized by hyperaldosteronism and high levels of 18-hydroxycortisol and 18-oxocortisol, which are under ACTH control.

REFERENCES

1. Fardella, C.E., et al. 2001. Genetic study of patients with dexamethasone-suppressible aldosteronism without the chimeric CYP11B1/CYP11B2 gene. J. Clin. Endocrinol. Metab. 86: 4805-4807.
2. Bechtel, S., et al. 2002. The effect of amino-acid substitutions I112P, D147E and K152N in CYP11B2 on the catalytic activities of the enzyme. Eur. J. Biochem. 269: 1118-1127.
3. Mukai, K., et al. 2003. An inverse correlation between expression of a preprocathepsin B-related protein with cysteine-rich sequences and steroid 11β-hydroxylase in adrenocortical cells. J. Biol. Chem. 278: 17084-17092.
4. Ganapathipillai, S., et al. 2005. CYP11B2-CYP11B1 haplotypes associated with decreased 11 β-hydroxylase activity. J. Clin. Endocrinol. Metab. 90: 1220-1225.
5. Krone, N., et al. 2005. Congenital adrenal hyperplasia due to 11-hydroxylase deficiency: functional characterization of two novel point mutations and a three-base pair deletion in the CYP11B1 gene. J. Clin. Endocrinol. Metab. 90: 3724-3730.
6. Barr, M., et al. 2006. Functional effects of genetic variants in the 11β-hydroxylase (CYP11B1) gene. Clin. Endocrinol. 65: 816-825.

CHROMOSOMAL LOCATION

Genetic locus: CYP11B1 (human) mapping to 8q24.3.

PRODUCT

CYP11B1 siRNA (h) is a pool of 3 target-specific 19-25 nt siRNAs designed to knock down gene expression. Each vial contains 3 nmol of lyophilized siRNA, sufficient for a 10 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CYP11B1 shRNA Plasmid (h): sc-44795-SH and CYP11B1 shRNA (h) Lentiviral Particles: sc-44795-V as alternate gene silencing products.

For independent verification of CYP11B1 (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-44795A, sc-44795B and sc-44795C.

RESEARCH USE

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

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

CYP11B1 siRNA (h) is recommended for the inhibition of CYP11B1 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

CYP11B1 (H-11): sc-374096 is recommended as a control antibody for monitoring of CYP11B1 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-IgGκ BP-HRP: sc-516102 or m-IgGκ 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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 CYP11B1 gene expression knockdown using RT-PCR Primer: CYP11B1 (h) -PR: sc-44795-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

PROTOCOLS

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