SANTA CRUZ BIOTECHNOLOGY, INC.

CCT A siRNA (h): sc-40394

BACKGROUND

Increase in fetal surfactant synthesis and lung maturity is caused by the glucocorticoidal induction of enzymes required for phosphatidylcholine synthesis towards the end of gestation. The regulation of gestational age-dependent induction of phosphatidylcholine synthesis by glucocorticoids is still unclear. The rate-controlling enzyme in the phosphatidylcholine biosynthetic pathway is CTP-phosphocholine cytidylyltransferase A (CCT A). In cultured eukaryotic cells, this enzyme is essential for survival. The alpha isoform is located in the nucleus and is regulated by reversible phosphorylation and membrane association. There is significant identity between the α-helical membrane-binding domains of CCT A and soybean oleosin. Expressed CCT A has lipid-dependent cytidylyltransferase activity. The gene which encodes CCT A maps to human chromosome 3q.

REFERENCES

1. Rutherford, M.S., et al. 1993. The gene for murine CTP: phosphocholine cytidylyltransferase (Ctpct) is located on mouse chromosome 16. Genomics 18: 698-701.
2. Hundertmark, S., et al. 1994. Gestational age dependence of 11 β-hydroxysteroid dehydrogenase and its relationship to the enzymes of phosphatidylcholine synthesis in lung and liver of fetal rat. Biochim. Biophys. Acta 1210: 348-354.
3. Kalmar, G.B., et al. 1994. Primary structure and expression of a human CTP:phosphocholine cytidylyltransferase. Biochim. Biophys. Acta 1219: 328-334.
4. Clement, J.M. and Kent, C. 1999. CTP: phosphocholine cytidylyltransferase: insights into regulatory mechanisms and novel functions. Biochem. Biophys. Res. Commun. 257: 643-650.
5. Online Mendelian Inheritance in Man, OMIM™. 2002. Johns Hopkins University, Baltimore, MD. MIM Number: 123695. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/

CHROMOSOMAL LOCATION

Genetic locus: PCYT1A (human) mapping to 3q29.

PRODUCT

CCT A 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see CCT A shRNA Plasmid (h): sc-40394-SH and CCT A shRNA (h) Lentiviral Particles: sc-40394-V as alternate gene silencing products.

For independent verification of CCT A (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-40394A, sc-40394B and sc-40394C.

PROTOCOLS

See our web site at www.scbt.com 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

CCT A siRNA (h) is recommended for the inhibition of CCT A 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

CCT A (F-6): sc-376107 is recommended as a control antibody for monitoring of CCT A 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 CCT A gene expression knockdown using RT-PCR Primer: CCT A (h)-PR: sc-40394-PR (20 µl). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

RESEARCH USE

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