SANTA CRUZ BIOTECHNOLOGY, INC.

citrin siRNA (h): sc-89601

BACKGROUND

Citrin, also known as SLC25A13 (solute carrier family 25 member 13), ARALAR2 or CTLN2, is a 675 amino acid multi-pass membrane protein that localizes to the inner membrane of the mitochondrion. Expressed in liver, pancreas, kidney, brain, heart and placenta, citrin functions as a calcium-dependent glutamate and aspartate carrier that is thought to play a role in the urea cycle. Citrin, a member of the mitochondrial carrier family, contains three Solcar repeats and four EF-hand domains through which it binds calcium. Defects in the gene encoding citrin are the cause of citrullinemia type 2 (CTLN2) and neonatal intrahepatic cholestasis due to citrin deficiency (NICCD). CTLN2 is an autosomal recessive disease that results from errors in the urea cycle and is characterized by neuropsychiatric symptoms such as loss of memory, seizures and coma. NICCD, a non-lethal disorder, occurs during infancy and is characterized by low birth weight, reduced bile flow, growth retardation and hepatic fibrosis.

REFERENCES

1. Sinasac, D.S., et al. 1999. Genomic structure of the adult-onset type II citrullinemia gene, SLC25A13, and cloning and expression of its mouse homologue. Genomics 62: 289-292.
2. Kobayashi, K., et al. 1999. The gene mutated in adult-onset type II citrullinaemia encodes a putative mitochondrial carrier protein. Nat. Genet. 22: 159-163.
3. Del Arco, A., et al. 2000. Characterization of a second member of the subfamily of calcium-binding mitochondrial carriers expressed in human non-excitable tissues. Biochem. J. 345: 725-732.
4. Palmieri, L., et al. 2001. Citrin and ARALAR1 are Ca^{2+}-stimulated aspartate/glutamate transporters in mitochondria. EMBO J. 20: 5060-5069.
5. Yamaguchi, N., et al. 2002. Screening of SLC25A13 mutations in early and late onset patients with citrin deficiency and in the Japanese population: Identification of two novel mutations and establishment of multiple DNA diagnosis methods for nine mutations. Hum. Mutat. 19: 122-130.
6. Saheki, T., et al. 2002. Mitochondrial aspartate glutamate carrier (citrin) deficiency as the cause of adult-onset type II citrullinemia (CTLN2) and idiopathic neonatal hepatitis (NICCD). J. Hum. Genet. 47: 333-341.

CHROMOSOMAL LOCATION

Genetic locus: SLC25A13 (human) mapping to 7q21.3.

PRODUCT

citrin 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 citrin shRNA Plasmid (h): sc-89601-SH and citrin shRNA (h) Lentiviral Particles: sc-89601-V as alternate gene silencing products.

For independent verification of citrin (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-89601A, sc-89601B and sc-89601C.

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

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

citrin (D-7): sc-393303 is recommended as a control antibody for monitoring of citrin 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 citrin gene expression knockdown using RT-PCR Primer: citrin (h)-PR: sc-89601-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.

PROTOCOLS

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