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ASS1 siRNA (h): sc-45810

BACKGROUND

ASS1, also known as argininosuccinate synthase or citrulline-aspartate ligase, belongs to the argininosuccinate synthase family. ASS1 is an urea cycle enzyme with a tetrameric structure composed of identical subunits. It is important to the urea cycle as it catalyzes the important second last step in the arginine biosynthetic pathway. A deficiency of ASS1 causes citrullinemia (CTLN1), an autosomal recessive disease which is characterized by severe vomiting spells and mental retardation.

REFERENCES

1. Bock, H.G., et al. 1983. Sequence for human argininosuccinate synthetase cDNA. *Nucleic Acids Res.* 11: 6505-6512.
2. Freytag, S.O., et al. 1984. Molecular structures of human argininosuccinate synthetase pseudogenes. Evolutionary and mechanistic implications. *J. Biol. Chem.* 259: 3160-3166.
3. Isashiki, Y., et al. 1989. Identification of essential arginine residue(s) for Mg-ATP binding of human argininosuccinate synthetase. *Protein Seq. Data Anal.* 2: 283-287.
4. Haberle, J., et al. 2002. Structure of the human argininosuccinate synthetase gene and an improved system for molecular diagnostics in patients with classical and mild citrullinemia. *Hum. Genet.* 110: 327-333.
5. Bansal, V., et al. 2004. Citrulline can preserve proliferation and prevent the loss of CD3 ζ chain under conditions of low arginine. *JPEN. J. Parenter. Enteral Nutr.* 28: 423-430.
6. Hao, G., et al. 2004. Argininosuccinate synthetase is reversibly inactivated by S-nitrosylation *in vitro* and *in vivo*. *J. Biol. Chem.* 279: 36192-36200.
7. Ito, S., et al. 2004. A pregnant patient with fulminant hepatic failure was found to carry a novel missense mutation in the argininosuccinate synthetase gene. *J. Gastroenterol.* 39: 1115-1117.
8. Lighthall, G.K., et al. 2004. Identification of salt-sensitive genes in the kidneys of Dahl rats. *J. Hypertens.* 22: 1487-1494.

CHROMOSOMAL LOCATION

Genetic locus: ASS1 (human) mapping to 9q34.11.

PRODUCT

ASS1 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 μ M solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see ASS1 shRNA Plasmid (h): sc-45810-SH and ASS1 shRNA (h) Lentiviral Particles: sc-45810-V as alternate gene silencing products.

For independent verification of ASS1 (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-45810A, sc-45810B and sc-45810C.

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 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

ASS1 siRNA (h) is recommended for the inhibition of ASS1 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

ASS1 (E-12): sc-365475 is recommended as a control antibody for monitoring of ASS1 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 ASS1 gene expression knockdown using RT-PCR Primer: ASS1 (h)-PR: sc-45810-PR (20 μ l, 599 bp). 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.