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ASS1 shRNA (h) Lentiviral Particles: sc-45810-V

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.
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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.
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CHROMOSOMAL LOCATION

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

PRODUCT

ASS1 shRNA (h) Lentiviral Particles is a pool of concentrated, transduction-ready viral particles containing 3 target-specific constructs that encode 19-25 nt (plus hairpin) shRNA designed to knock down gene expression. Each vial contains 200 μ l frozen stock containing 1.0×10^6 infectious units of virus (IFU) in Dulbecco's Modified Eagle's Medium with 25 mM HEPES pH 7.3. Suitable for 10-20 transductions. Also see ASS1 siRNA (h): sc-45810 and ASS1 shRNA Plasmid (h): sc-45810-SH as alternate gene silencing products.

STORAGE

Store lentiviral particles at -80° C. Stable for at least one year from the date of shipment. Once thawed, particles can be stored at 4° C for up to one week. Avoid repeated freeze thaw cycles.

APPLICATIONS

ASS1 shRNA (h) Lentiviral Particles is recommended for the inhibition of ASS1 expression in human cells.

SUPPORT REAGENTS

Control shRNA Lentiviral Particles: sc-108080. Available as 200 μ l frozen viral stock containing 1.0×10^6 infectious units of virus (IFU); contains an shRNA construct encoding a scrambled sequence that will not lead to the specific degradation of any known cellular mRNA.

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 (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 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 $55-60^\circ$ C and the extension temperature should be $68-72^\circ$ C.

BIOSAFETY

Lentiviral particles can be employed in standard Biosafety Level 2 tissue culture facilities (and should be treated with the same level of caution as with any other potentially infectious reagent). Lentiviral particles are replication-incompetent and are designed to self-inactivate after transduction and integration of shRNA constructs into genomic DNA of target cells.

RESEARCH USE

The purchase of this product conveys to the buyer the nontransferable right to use the purchased amount of the product and all replicates and derivatives for research purposes conducted by the buyer in his laboratory only (whether the buyer is an academic or for-profit entity). The buyer cannot sell or otherwise transfer (a) this product (b) its components or (c) materials made using this product or its components to a third party, or otherwise use this product or its components or materials made using this product or its components for Commercial Purposes.

PROTOCOLS

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