SANTA CRUZ BIOTECHNOLOGY, INC.

ALDH18A1 siRNA (m): sc-140997

BACKGROUND

Aldehyde dehydrogenases (ALDHs) mediate $NADP^+$-dependent oxidation of aldehydes into acids during the detoxification of alcohol-derived acetaldehyde, lipid peroxidation, and metabolism of corticosteroids, biogenic amines and neurotransmitters. ALDH18A1 (aldehyde dehydrogenase 18 family member A1), also known as GSAS (glutamate γ-semialdehyde synthetase), P5CS (δ1-pyrroline-5-carboxylate synthetase) or PYCS, is the major enzyme involved in proline, arginine and ornithine biosynthesis. Localizing to the inner mitochondrial membrane, ALDH18A1 catalyzes the reduction of glutamate to δ1-pyrroline-5-carboxylate. Due to alternative splicing events, two isoforms exist for ALDH18A1, differing by only two amino acids. The longer isoform is widely expressed while the shorter isoform predominantly functions in the gut. A mutation in the gene encoding ALDH18A1 results in P5CS deficiency, a disease characterized by progressive joint laxity, neurodegeneration, bilateral subcapsular cataracts and skin hyperelasticity.

REFERENCES

1. Jones, C. 1975. Synteny between the pro^+ marker and human glutamate oxaloacetate transaminase. Somatic Cell Genet. 1: 345-354.
2. Liu, G., et al. 1996. Assignment of the human gene encoding the δ1-pyrroline-5-carboxylate synthetase (P5CS) to 10q24.3 by *in situ* hybridization. Genomics 37: 145-146.
3. Aral, B., et al. 1996. Database cloning human δ1-pyrroline-5-carboxylate synthetase (P5CS) cDNA: a bifunctional enzyme catalyzing the first 2 steps in proline biosynthesis. C. R. Acad. Sci. III, Sci. Vie 319: 171-178.
4. Hu, C.A., et al. 1999. Molecular enzymology of mammalian δ1-pyrroline-5-carboxylate synthase. Alternative splice donor utilization generates isoforms with different sensitivity to ornithine inhibition. J. Biol. Chem. 274: 6754-6762.

CHROMOSOMAL LOCATION

Genetic locus: Aldh18a1 (mouse) mapping to 19 C3.

PRODUCT

ALDH18A1 siRNA (m) 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 ALDH18A1 shRNA Plasmid (m): sc-140997-SH and ALDH18A1 shRNA (m) Lentiviral Particles: sc-140997-V as alternate gene silencing products.

For independent verification of ALDH18A1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-140997A, sc-140997B and sc-140997C.

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

ALDH18A1 siRNA (m) is recommended for the inhibition of ALDH18A1 expression in mouse 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

ALDH18A1 (G-10): sc-515443 is recommended as a control antibody for monitoring of ALDH18A1 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 ALDH18A1 gene expression knockdown using RT-PCR Primer: ALDH18A1 (m)-PR: sc-140997-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.