

ACADSB siRNA (h): sc-90519

BACKGROUND

The Acyl-CoA dehydrogenase (ACAD) family of enzymes are involved in the catabolism of fatty acids and amino acids. They provide a major source of energy for the heart and skeletal muscle. The short/branched chain specific acyl-CoA dehydrogenase (ACADSB), also designated 2-methylbutyryl-coenzyme A dehydrogenase, is a 432 amino acid protein that is ubiquitously expressed. Specifically, ACADSB forms a homotetramer within the mitochondrial matrix. ACADSB catalyzes the degradation of L-isoleucine and has the highest affinity for (s)-2-methylbutyryl-CoA, isobutyryl-CoA and 2-methylhexanoyl-CoA as substrates. Mutations in the gene encoding ACADSB result in Defects in ACADSB are the cause of short/branched-chain acyl-CoA dehydrogenase deficiency (SBCADD), an autosomal recessive disorder characterized by an increase of 2-methylbutyrylglycine and 2-methylbutyrylcarnitine in blood and urine. Patients with SBCADD have seizures and psychomotor delay as the main clinical features.

REFERENCES

1. Rozen, R., Vockley, J., Zhou, L., Milos, R., Willard, J., Fu, K., Vicanek, C., Low-Nang, L., Torban, E. and Fournier, B. 1994. Isolation and expression of a cDNA encoding the precursor for a novel member (ACADSB) of the acyl-CoA dehydrogenase gene family. *Genomics* 24: 280-287.
2. Arden, K.C., Viars, C.S., Fu, K. and Rozen, R. 1995. Localization of short/branched chain acyl-CoA dehydrogenase (ACADSB) to human chromosome 10. *Genomics* 25: 743-745.
3. Korman, S.H., Andresen, B.S., Zeharia, A., Gutman, A., Boneh, A. and Pitt, J.J. 2005. 2-ethylhydracrylic aciduria in short/branched-chain acyl-CoA dehydrogenase deficiency: application to diagnosis and implications for the R-pathway of isoleucine oxidation. *Clin. Chem.* 51: 610-617.
4. Korman, S.H. 2006. Inborn errors of isoleucine degradation: a review. *Mol. Genet. Metab.* 89: 289-299.

CHROMOSOMAL LOCATION

Genetic locus: ACADSB (human) mapping to 10q26.13.

PRODUCT

ACADSB 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 ACADSB shRNA Plasmid (h): sc-90519-SH and ACADSB shRNA (h) Lentiviral Particles: sc-90519-V as alternate gene silencing products.

For independent verification of ACADSB (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-90519A, sc-90519B and sc-90519C.

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 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

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

ACADSB (C-9): sc-398773 is recommended as a control antibody for monitoring of ACADSB 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 ACADSB gene expression knockdown using RT-PCR Primer: ACADSB (h)-PR: sc-90519-PR (20 μ l). 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.