

DCAKD siRNA (h): sc-93599

BACKGROUND

Coenzyme A (CoA) is an essential cofactor used in numerous biochemical pathways. It plays a critical role in the synthesis and oxidation of fatty acids and is vital to the citric acid cycle. The biosynthesis pathway of CoA from pantothenic acid (also known as vitamin B₅) is essential and universal in prokaryotes and eukaryotes. In humans, the final steps of the biosynthesis pathway are carried out by the bifunctional enzyme COASY. The sequence of these enzymes are highly conserved between different bacterial species. The phosphopantetheine adenylyltransferase and dephospho-coenzyme A kinase activities of COASY are evolutionarily conserved activities. DCAKD (dephospho-CoA kinase domain containing protein) is a 231 amino acid protein that consists of a dephospho-CoA kinase domain and an ATP nucleotide binding motif. Localizing to mitochondria and the cytosol, DCAKD belongs to the coaE family which suggests that it may play a role in the biosynthesis of CoA.

REFERENCES

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- Aghajanian, S. and Worrall, D.M. 2002. Identification and characterization of the gene encoding the human phosphopantetheine adenylyltransferase and dephospho-CoA kinase bifunctional enzyme (CoA synthase). *Biochem. J.* 365: 13-18.
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CHROMOSOMAL LOCATION

Genetic locus: DCAKD (human) mapping to 17q21.31.

PRODUCT

DCAKD 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 DCAKD shRNA Plasmid (h): sc-93599-SH and DCAKD shRNA (h) Lentiviral Particles: sc-93599-V as alternate gene silencing products.

For independent verification of DCAKD (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-93599A, sc-93599B and sc-93599C.

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

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

DCAKD (SQ-14): sc-100348 is recommended as a control antibody for monitoring of DCAKD 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 DCAKD gene expression knockdown using RT-PCR Primer: DCAKD (h)-PR: sc-93599-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.

PROTOCOLS

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