SANTA CRUZ BIOTECHNOLOGY, INC.

COASY siRNA (m): sc-142448

BACKGROUND

COASY (coenzyme A synthase), also known as NBP (nucleotide binding protein), DPCK (dephospho-coenzyme A kinase), PPAT (pantetheine-phosphate adenylyltransferase), UKR1 or pOV-2, is a bifunctional enzyme involved in the biosynthesis of coenzyme A (CoA). COASY exhibits both Ppat activity and DPCK activity, catalyzing steps four and five, respectively, of the CoA biosynthetic pathway. Functioning as a widely expressed monomer and induced by phospholipids, COASY localizes to the outer mitochondrial membrane and facilitates the conversion of 4'-phosphopantetheine to dephospho-CoA and the subsequent generation of CoA. CoA is an important molecule in the cell, participating in carbohydrate, amino acid and fatty acid metabolism. It is the predominant acetyl and acyl group carrier and is used as a substrate by approximately 4% of all cellular enzymes. Due to alternative splicing events, an additional isoform of COASY, namely COASY b, is expressed in brain and contains an extra 29 amino acids at the N-terminus.

REFERENCES

1. 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.
2. Daugherty, M., et al. 2002. Complete reconstitution of the human coenzyme A biosynthetic pathway via comparative genomics. J. Biol. Chem. 277: 21431-21439.
3. Zhyvoloup, A., et al. 2002. Molecular cloning of CoA synthase. The missing link in CoA biosynthesis. J. Biol. Chem. 277: 22107-22110.
4. Zhyvoloup, A., et al. 2003. Subcellular localization and regulation of coenzyme A synthase. J. Biol. Chem. 278: 50316-50321.
5. Nemazanyy, I., et al. 2004. Specific interaction between S6K1 and CoA synthase: a potential link between the mTOR/S6K pathway, CoA biosynthesis and energy metabolism. FEBS Lett. 578: 357-362.
6. Kardia, S.R., et al. 2006. Characterizing variation in sex steroid hormone pathway genes in women of 4 races/ethnicities: the Study of Women's Health Across the Nation (SWAN). Am. J. Med. 119: S3-S15.

CHROMOSOMAL LOCATION

Genetic locus: Coasy (mouse) mapping to 11 D.

PRODUCT

COASY 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 COASY shRNA Plasmid (m): sc-142448-SH and COASY shRNA (m) Lentiviral Particles: sc-142448-V as alternate gene silencing products.

For independent verification of COASY (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-142448A, sc-142448B and sc-142448C.

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

COASY siRNA (m) is recommended for the inhibition of COASY 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

COASY (E-7): sc-393812 is recommended as a control antibody for monitoring of COASY 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 COASY gene expression knockdown using RT-PCR Primer: COASY (m)-PR: sc-142448-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.

PROTOCOLS

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