SANTA CRUZ BIOTECHNOLOGY, INC.

PHACS siRNA (h): sc-62792

BACKGROUND

PHACS (putative human ACS), also known as ACC or ACS (1-aminocyclopropane-1-carboxylate synthase homolog), is a 501 amino acid protein that belongs to the α family of pyridoxal-5'-phosphate enzymes. PHACS is expressed in a wide range of tissues and shares structural similarity with AAT (aspartate aminotransferase), TAT (tyrosine aminotransferase) and enzymes that catalyze β-elimination reactions on amino acids. PHACS consists of overlapping aminotransferase I and β-eliminating lyase domains and is involved in the deamination of L-vinylglycine. The plant homolog ACS is the key metabolic intermediate in the biosynthesis of phytohormone ethylene, which is essential for the growth and development of plants. Unlike ACS, PHACS does not catalyze the synthesis of 1-aminocyclopropane-1-carboxylate.

REFERENCES

1. Penrose, D.M. and Glick, B.R. 1997. Enzymes that regulate ethylene levels—1-aminocyclopropane-1-carboxylic acid (ACC) deaminase, ACC synthase and ACC oxidase. Indian J. Exp. Biol. 35: 1-17.
2. Capitani, G., et al. 1999. Structure of 1-aminocyclopropane-1-carboxylate synthase, a key enzyme in the biosynthesis of the plant hormone ethylene. J. Mol. Biol. 294: 745-756.
3. Feng, L. and Kirsch, J.F. 2000. L-Vinylglycine is an alternative substrate as well as a mechanism-based inhibitor of 1-aminocyclopropane-1-carboxylate synthase. Biochemistry 39: 2436-2444.
4. Feng, L., et al. 2000. Aminotransferase activity and bioinformatic analysis of 1-aminocyclopropane-1-carboxylate synthase. Biochemistry 39: 15242-15249.
5. Peixoto, B.R., et al. 2000. Characterization of the recombinase activating gene-1 and 2 locus in the Japanese pufferfish, Fugu rubripes. Gene 246: 275-283.
6. Koch, K.A., et al. 2001. The human cDNA for a homologue of the plant enzyme 1-aminocyclopropane-1-carboxylate synthase encodes a protein lacking that activity. Gene 272: 75-84.

CHROMOSOMAL LOCATION

Genetic locus: ACCS (human) mapping to 11p11.2.

PRODUCT

PHACS 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see PHACS shRNA Plasmid (h): sc-62792-SH and PHACS shRNA (h) Lentiviral Particles: sc-62792-V as alternate gene silencing products.

For independent verification of PHACS (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-62792A, sc-62792B and sc-62792C.

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

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

PHACS (MA-02): sc-130477 is recommended as a control antibody for monitoring of PHACS 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 PHACS gene expression knockdown using RT-PCR Primer: PHACS (h)-PR: sc-62792-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.