SANTA CRUZ BIOTECHNOLOGY, INC.

PDHA2 siRNA (m): sc-106393

BACKGROUND

The pyruvate dehydrogenase (PDH) complex is a nuclear-encoded mitochondrial matrix enzyme complex that functions as the primary link between glycolysis and the tricarboxylic acid (TCA) cycle by catalyzing the irreversible conversion of pyruvate into acetyl-CoA, an essential step in aerobic glucose metabolism. PDHA2 (pyruvate dehydrogenase α 2), also known as PDHAL, is a 388 amino acid mitochondrial matrix protein expressed in postmeiotic spermatogenic cells. Composed of a tetramer containing two α and two β subunits, PDHA2 consists multiple copies of three enzymatic components: pyruvate dehydrogenase (E1), dihydrolipoamide acetyltransferase (E2) and lipoamide dehydrogenase (E3). PHDA2 is suggested to participate it in cell proliferation and may be involved in prostate cancer. PHDA2 is encoded by a gene located on human chromosome 4, which encodes nearly 6% of the human genome and has the largest gene deserts (regions of the genome with no protein encoding genes) of all of the human chromosomes.

REFERENCES

1. Dahl, H.H., et al. 1990. A testis-specific form of the human pyruvate dehydrogenase E1 α subunit is coded for by an intronless gene on chromosome 4. Genomics 8: 225-232.
2. Brown, R.M., et al. 1990. Pyruvate dehydrogenase E1 α subunit genes in the mouse: mapping and comparison with human homologs. Somat. Cell Mol. Genet. 16: 487-492.
3. Online Mendelian Inheritance in Man, OMIM™. 1990. Johns Hopkins University, Baltimore, MD. MIM Number:179061. World Wide Web URL: http://www.ncbi.nlm.nih.gov/omim/
4. Fitzgerald, J., et al. 1992. Isolation and characterisation of the mouse pyruvate dehydrogenase E1 α genes. Biochim. Biophys. Acta 1131: 83-90.
5. Lanneluc, I., et al. 1996. Synteny conservation between parts of human chromosome 4q and bovine and ovine chromosomes 6. Cytogenet. Cell Genet. 72: 212-214.
6. Klar, J., et al. 2005. RAR-related orphan receptor A isoform 1 (RORa1) is disrupted by a balanced translocation t(4;15)(q22.3;q21.3) associated with severe obesity. Eur. J. Hum. Genet. 13: 928-934.

CHROMOSOMAL LOCATION

Genetic locus: Pdha2 (mouse) mapping to 3 H1.

PRODUCT

PDHA2 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 PDHA2 shRNA Plasmid (m): sc-106393-SH and PDHA2 shRNA (m) Lentiviral Particles: sc-106393-V as alternate gene silencing products.

For independent verification of PDHA2 (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-106393A, sc-106393B and sc-106393C.

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

PDHA2 siRNA (m) is recommended for the inhibition of PDHA2 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

PDHA2 (B-3): sc-393219 is recommended as a control antibody for monitoring of PDHA2 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 PDHA2 gene expression knockdown using RT-PCR Primer: PDHA2 (m)-PR: sc-106393-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.