

CLS1 siRNA (h): sc-72929

BACKGROUND

Cardiolipin synthetase, CLS1, is a mitochondrial protein that belongs to the CDP-alcohol phosphatidyltransferase class-I family. CLS1 is a multi-pass membrane protein localized to the inner membrane of mitochondria. CLS1 is responsible for the catalyzing the reversible transfer of a phosphatidyl group from one phosphatidyl glycerol molecule to another. This process results in the formation of cardiolipin (CL, or diphosphatidyl glycerol) and glycerol. Diphosphatidyl glycerol is a major component of the mitochondrial membrane and constitutes roughly 20% of total mitochondrial lipids. Having four fatty acid tails rather than the usual two, CL is a double phospholipid that is synthesized in the mitochondrion itself. Defects in the CLS1 gene are likely to effect metabolism, sustained production of ATP and could contribute to diseases such as Barth syndrome.

REFERENCES

1. Taylor, W.A., et al. 2002. Expression of monolysocardiolipin acyltransferase activity is regulated in concert with the level of cardiolipin and cardiolipin biosynthesis in the mammalian heart. *BMC Biochem.* 3: 9.
2. Zhong, Q., et al. 2004. Absence of cardiolipin results in temperature sensitivity, respiratory defects, and mitochondrial DNA instability independent of pet56. *J. Biol. Chem.* 279: 32294-32300.
3. Chen, D., et al. 2006. Identification and functional characterization of hCLS1, a human cardiolipin synthase localized in mitochondria. *Biochem. J.* 398: 169-176.
4. Houtkooper, R.H., et al. 2006. Identification and characterization of human cardiolipin synthase. *FEBS Lett.* 580: 3059-3064.
5. Lu, B., et al. 2006. Cloning and characterization of a cDNA encoding human cardiolipin synthase (hCLS1). *J. Lipid Res.* 47: 1140-1145.
6. Choi, S.Y., et al. 2007. Cardiolipin deficiency releases cytochrome c from the inner mitochondrial membrane and accelerates stimuli-elicited apoptosis. *Cell Death Differ.* 14: 597-606.

CHROMOSOMAL LOCATION

Genetic locus: CLRS1 (human) mapping to 20p12.3.

PRODUCT

CLS1 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 CLS1 shRNA Plasmid (h): sc-72929-SH and CLS1 shRNA (h) Lentiviral Particles: sc-72929-V as alternate gene silencing products.

For independent verification of CLS1 (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-72929A, sc-72929B and sc-72929C.

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

CLS1 siRNA (h) is recommended for the inhibition of CLS1 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

CLS1 (E-8): sc-514986 is recommended as a control antibody for monitoring of CLS1 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 CLS1 gene expression knockdown using RT-PCR Primer: CLS1 (h)-PR: sc-72929-PR (20 μ l, 400 bp). 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.