LCMT1 siRNA (h): sc-93344



The Power to Question

BACKGROUND

Protein phosphatase 2A (PP2A) is a serine/threonine (Ser/Thr) phosphatase that is thought to be involved in cell growth and proliferation events and may be associated with tumor progression. The activity of PP2A is regulated by a variety of mechanisms, one of which is the reversible methylation by select methyltransferases. LCMT1 (leucine carboxyl methyltransferase 1), also known as LCMT, PPMT1 or CGI-68, is a 334 amino acid member of the methyltransferase superfamily that is involved in the regulation of PP2A. Specifically, LCMT1 catalyzes the methylation of the carboxy group on the C-terminal leucine of the PP2A catalytic subunit (designated PP2A α). Via its ability to regulate PP2A function, LCMT1 may be critical for normal mitotic progression and overall cell survival. Two isoforms of LCMT1 are expressed due to alternative splicing events.

REFERENCES

- De Baere, I., Derua, R., Janssens, V., Van Hoof, C., Waelkens, E., Merlevede, W. and Goris, J. 1999. Purification of porcine brain protein phosphatase 2A leucine carboxyl methyltransferase and cloning of the human homologue. Biochemistry 38: 16539-16547.
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- Longin, S., Zwaenepoel, K., Louis, J.V., Dilworth, S., Goris, J. and Janssens, V. 2007. Selection of protein phosphatase 2A regulatory subunits is mediated by the C terminus of the catalytic subunit. J. Biol. Chem. 282: 26971-26980.
- Lee, J.A. and Pallas, D.C. 2007. Leucine carboxyl methyltransferase-1 is necessary for normal progression through mitosis in mammalian cells.
 J. Biol. Chem. 282: 30974-30984.

CHROMOSOMAL LOCATION

Genetic locus: LCMT1 (human) mapping to 16p12.1.

PRODUCT

LCMT1 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 LCMT1 shRNA Plasmid (h): sc-93344-SH and LCMT1 shRNA (h) Lentiviral Particles: sc-93344-V as alternate gene silencing products.

For independent verification of LCMT1 (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-93344A, sc-93344B and sc-93344C.

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

LCMT1 siRNA (h) is recommended for the inhibition of LCMT1 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

LCMT1 (D-10): sc-365221 is recommended as a control antibody for monitoring of LCMT1 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-lgG κ BP-HRP: sc-516102 or m-lgG κ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz MarkerTM Molecular Weight Standards: sc-2035, UltraCruz[®] Blocking Reagent: sc-516214 and Western Blotting Luminol Reagent: sc-2048. 2) Immunofluorescence: use m-lgG κ BP-FITC: sc-516140 or m-lgG 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 LCMT1 gene expression knockdown using RT-PCR Primer: LCMT1 (h)-PR: sc-93344-PR (20 μ I, 503 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.

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