

pICln siRNA (h): sc-42594

BACKGROUND

The formation of the spliceosome includes the assembly of Sm proteins in an ordered manner onto snRNAs. This process is mediated by the survival of motor neuron (SMN) protein, and is enhanced by modification of specific arginine residues in the Sm proteins to symmetrical dimethylarginines (sDMAs). sDMA modification of Sm proteins is catalyzed by the methylosome, a complex comprised of the type II methyltransferase PRMT5 (also designated Jak-binding protein 1, JBP1), pICln, and two novel factors. PRMT5 binds the Sm proteins via their arginine- and glycine-rich (RG) domains, while pICln binds the Sm domains. pICln also acts as an inhibitor of SnRNP assembly by preventing specific interactions between Sm proteins required for the formation of the Sm core. pICln is a highly conserved, ubiquitously expressed protein that localizes primarily to the cytoplasm, and may play a role as a swelling-activated anion channel or a channel regulator in addition to its function in the methylosome. The gene encoding human pICln maps to chromosome 11q14.1.

REFERENCES

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- Emma, F., et al. 1998. Characterization of pICln binding proteins: identification of p17 and assessment of the role of acidic domains in mediating protein-protein interactions. *Biochim. Biophys. Acta* 1404: 321-328.
- Li, C., et al. 1998. Recombinant pICln forms highly cation-selective channels when reconstituted into artificial and biological membranes. *J. Gen. Physiol.* 112: 727-736.
- Pu, W.T., et al. 2000. ICln is essential for cellular and early embryonic viability. *J. Biol. Chem.* 275: 12363-12366.
- Friesen, W.J., et al. 2001. The methylosome, a 20S complex containing JBP1 and pICln, produces dimethylarginine-modified Sm proteins. *Mol. Cell. Biol.* 21: 8289-8300.
- Meister, G., et al. 2001. Methylation of Sm proteins by a complex containing PRMT5 and the putative U snRNP assembly factor pICln. *Curr. Biol.* 11: 1990-1994.

CHROMOSOMAL LOCATION

Genetic locus: CLNS1A (human) mapping to 11q14.1.

PRODUCT

pICln 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 pICln shRNA Plasmid (h): sc-42594-SH and pICln shRNA (h) Lentiviral Particles: sc-42594-V as alternate gene silencing products.

For independent verification of pICln (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-42594A, sc-42594B and sc-42594C.

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

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

pICln (G-1): sc-271327 is recommended as a control antibody for monitoring of pICln 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 pICln gene expression knockdown using RT-PCR Primer: pICln (h)-PR: sc-42594-PR (20 μ l). 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.