

SLA/LP siRNA (h): sc-89108

BACKGROUND

The fidelity of protein synthesis requires efficient discrimination of amino acid substrates by aminoacyl-tRNA synthetases. Aminoacyl-tRNA synthetases function to catalyze the aminoacylation of tRNAs by their corresponding amino acids, thus linking amino acids with tRNA-contained nucleotide triplets. SLA/LP (soluble liver antigen/liver-pancreas antigen), also known as SEPSECS (Sep (O-phosphoserine) tRNA:Sec (selenocysteine) tRNA synthase) or SLA-p35, is a 501 amino acid cytoplasmic protein that belongs to a diverse family of pyridoxal phosphate-dependent enzymes. Expressed predominantly in liver, lung, kidney and pancreas, SLA/LP plays a role in aminoacyl-tRNA synthesis and, more specifically, selenoprotein biosynthesis. Using PLP as a cofactor, SLA/LP specifically converts O-phosphoseryl-tRNA^{Sec} to Sec-tRNA^{Sec} by exchanging the phosphate group for a selenol moiety. Due to alternative splicing events, two SLA/LP isoforms exist.

REFERENCES

- Costa, M., et al. 2000. Isolation and characterization of cDNA encoding the antigenic protein of the human tRNP^(Ser)^{Sec} complex recognized by autoantibodies from patients with type-1 autoimmune hepatitis. Clin. Exp. Immunol. 121: 364-374.
- Volkman, M., et al. 2001. Soluble liver antigen: isolation of a 35-kd recombinant protein (SLA-p35) specifically recognizing sera from patients with autoimmune hepatitis. Hepatology 33: 591-596.
- Xu, X.M., et al. 2005. Evidence for direct roles of two additional factors, SECp43 and soluble liver antigen, in the selenoprotein synthesis machinery. J. Biol. Chem. 280: 41568-41575.
- Yuan, J., et al. 2006. RNA-dependent conversion of phosphoserine forms selenocysteine in eukaryotes and archaea. Proc. Natl. Acad. Sci. USA 103: 18923-18927.
- Xu, X.M., et al. 2007. Biosynthesis of selenocysteine on its tRNA in eukaryotes. PLoS Biol. 5: e4.
- Hauenstein, S.I. and Perona, J.J. 2008. Redundant synthesis of cysteinyl-tRNA^{Cys} in *Methanosarcina mazei*. J. Biol. Chem. 283: 22007-22017.
- Araiso, Y., et al. 2008. Structural insights into RNA-dependent eukaryal and archaeal selenocysteine formation. Nucleic Acids Res. 36: 1187-1199.

CHROMOSOMAL LOCATION

Genetic locus: SEPSECS (human) mapping to 4p15.2.

PRODUCT

SLA/LP 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 SLA/LP shRNA Plasmid (h): sc-89108-SH and SLA/LP shRNA (h) Lentiviral Particles: sc-89108-V as alternate gene silencing products.

For independent verification of SLA/LP (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-89108A, sc-89108B and sc-89108C.

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

SLA/LP siRNA (h) is recommended for the inhibition of SLA/LP 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

SLA/LP (E-11): sc-514729 is recommended as a control antibody for monitoring of SLA/LP 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 MarkerTM 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 SLA/LP gene expression knockdown using RT-PCR Primer: SLA/LP (h)-PR: sc-89108-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.