

Asparagine synthetase siRNA (h): sc-60212

BACKGROUND

Glutamine-hydrolyzing Asparagine synthetase is also commonly designated cell cycle control protein TS11. Asparagine synthetase plays an important role in the amino-acid biosynthesis pathway and is also important for L-asparagine biosynthesis. Via the L-glutamine route, it is involved in the synthesis of L-asparagine from L-aspartate. The protein contains one Asparagine synthetase domain and one type-2 glutamine amidotransferase domain. The cell-cycle regulated gene encoding for Asparagine synthetase, *ts11*, is necessary for G₁ progression.

REFERENCES

1. Andrulis, I.L., et al. 1987. Isolation of human cDNAs for Asparagine synthetase and expression in Jensen rat sarcoma cells. *Mol. Cell. Biol.* 7: 2435-2443.
2. Van Heeke, G., et al. 1989. The N-terminal cysteine of human Asparagine synthetase is essential for glutamine-dependent activity. *J. Biol. Chem.* 264: 19475-19477.
3. Greco, A., et al. 1989. Organization and expression of the cell cycle gene, *ts11*, that encodes Asparagine synthetase. *Mol. Cell. Biol.* 9: 2350-2359.
4. Chen, H., et al. 2004. Amino acid deprivation induces the transcription rate of the human Asparagine synthetase gene through a timed program of expression and promoter binding of nutrient-responsive basic region/leucine zipper transcription factors as well as localized histone acetylation. *J. Biol. Chem.* 279: 50829-50839.

CHROMOSOMAL LOCATION

Genetic locus: *ASNS* (human) mapping to 7q21.3.

PRODUCT

Asparagine synthetase 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 Asparagine synthetase shRNA Plasmid (h): sc-60212-SH and Asparagine synthetase shRNA (h) Lentiviral Particles: sc-60212-V as alternate gene silencing products.

For independent verification of Asparagine synthetase (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-60212A, sc-60212B and sc-60212C.

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

Asparagine synthetase siRNA (h) is recommended for the inhibition of Asparagine synthetase 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

Asparagine synthetase (G-10): sc-365809 is recommended as a control antibody for monitoring of Asparagine synthetase 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 Asparagine synthetase gene expression knockdown using RT-PCR Primer: Asparagine synthetase (h)-PR: sc-60212-PR (20 μ l, 442 bp). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Linares, J.F., et al. 2017. ATF4-induced metabolic reprogramming is a synthetic vulnerability of the p62-deficient tumor stroma. *Cell Metab.* 26: 817-829.e6.
2. Cui, J., et al. 2023. Methylseleninic acid overcomes gefitinib resistance through asparagine-MET-TOPK signaling axis in non-small cell lung cancer cells. *Biochem. Pharmacol.* 215: 115690.

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.