

CPSF3 siRNA (h): sc-105243

BACKGROUND

3' ends of eukaryotic mRNAs can undergo processing events that include endonucleolytic cleavage and polyadenylation. Cleavage and polyadenylation specificity factor (CPSF) mediates 3' cleavage of the transcript and subsequent polyadenylation. CPSF contains four subunits and localizes to the nucleoplasm where it recognizes the AAUAAA signal in pre-mRNA and interacts with other proteins to facilitate RNA cleavage and poly(A) synthesis. The human CPSF3 gene maps to chromosome 2p25.1 and encodes the second shortest subunit of cleavage and polyadenylation specificity factor. U1 snRNP-A protein (U1A) interacts with and affects the activity of CPSF by stabilizing the interaction of CPSF with the AAUAAA-containing RNAs to increase the efficiency of polyadenylation.

REFERENCES

1. Murthy, K.G. and Manley, J.L. 1995. The 160-kD subunit of human cleavage-polyadenylation specificity factor coordinates pre-mRNA 3'-end formation. *Genes Dev.* 9: 2672-2683.
2. Lutz, C.S., et al. 1996. Interaction between the U1 snRNP-A protein and the 160-kD subunit of cleavage-polyadenylation specificity factor increases polyadenylation efficiency *in vitro*. *Genes Dev.* 10: 325-337.
3. Jenny, A., et al. 1996. Sequence similarity between the 73-kilodalton protein of mammalian CPSF and a subunit of yeast polyadenylation factor I. *Science* 274: 1514-1517.
4. Barabino, S.M., et al. 1997. The 30-kD subunit of mammalian cleavage and polyadenylation specificity factor and its yeast homolog are RNA-binding zinc finger proteins. *Genes Dev.* 11: 1703-1716.
5. Salinas, C.A., et al. 1998. Characterization of a *Drosophila* homologue of the 160-kDa subunit of the cleavage and polyadenylation specificity factor CPSF. *Mol. Gen. Genet.* 257: 672-680.
6. Edmonds, M. 2002. A history of poly A sequences: from formation to factors to function. *Prog. Nucleic Acid Res. Mol. Biol.* 71: 285-389.

CHROMOSOMAL LOCATION

Genetic locus: CPSF3 (human) mapping to 2p25.1.

PRODUCT

CPSF3 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 CPSF3 shRNA Plasmid (h): sc-105243-SH and CPSF3 shRNA (h) Lentiviral Particles: sc-105243-V as alternate gene silencing products.

For independent verification of CPSF3 (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-105243A, sc-105243B and sc-105243C.

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

CPSF3 siRNA (h) is recommended for the inhibition of CPSF3 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

CPSF3 (C-3): sc-393001 is recommended as a control antibody for monitoring of CPSF3 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 CPSF3 gene expression knockdown using RT-PCR Primer: CPSF3 (h)-PR: sc-105243-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.