

CPSF6 siRNA (h): sc-72990

BACKGROUND

3' ends of eukaryotic mRNAs can undergo processing events that include endonucleolytic cleavage and polyadenylation. Cleavage and polyadenylation specificity factors (CPSF) mediate 3' cleavage of the transcript and subsequent polyadenylation. CPSF6, also known as CFIm68 (mammalian cleavage factor I, 68 kDa subunit), HPBR11-4 or HPBR11-7, is a member of the CPSF6/7 family and contains an N-terminal RNA recognition motif (RRM) and a C-terminal RS-like domain. Via its RS-like domain, CPSF6 interacts with SRp20, Tra-2 β and 9G8. CPSF6 localizes to the paraspeckles and forms a heterodimer with NUDT21, comprising the CFIm complex which is essential for the first step in pre-mRNA 3' cleavage and polyadenylation processing. CPSF6 is the larger subunit of the complex and is present in only half of the two heterodimer combinations (the other half being a dimer of NUDT21 and CPSF7).

REFERENCES

1. 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.
2. 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.
3. Edmonds, M. 2002. A history of poly A sequences: from formation to factors to function. *Prog. Nucleic Acid Res. Mol. Biol.* 71: 285-389.
4. Dettwiler, S., et al. 2004. Distinct sequence motifs within the 68-kDa subunit of cleavage factor Im mediate RNA binding, protein-protein interactions, and subcellular localization. *J. Biol. Chem.* 279: 35788-35797.
5. López-Camarillo, C., et al. 2005. *Entamoeba histolytica*: comparative genomics of the pre-mRNA 3' end processing machinery. *Exp. Parasitol.* 110: 184-190.
6. Millevoi, S., et al. 2006. An interaction between U2AF65 and CF Im links the splicing and 3' end processing machineries. *EMBO J.* 25: 4854-4864.

CHROMOSOMAL LOCATION

Genetic locus: CPSF6 (human) mapping to 12q15.

PRODUCT

CPSF6 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 CPSF6 shRNA Plasmid (h): sc-72990-SH and CPSF6 shRNA (h) Lentiviral Particles: sc-72990-V as alternate gene silencing products.

For independent verification of CPSF6 (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-72990A, sc-72990B and sc-72990C.

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

CPSF6 siRNA (h) is recommended for the inhibition of CPSF6 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

CPSF6 (F-3): sc-376228 is recommended as a control antibody for monitoring of CPSF6 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 CPSF6 gene expression knockdown using RT-PCR Primer: CPSF6 (h)-PR: sc-72990-PR (20 μ l). Annealing temperature for the primers should be 55-60° C and the extension temperature should be 68-72° C.

SELECT PRODUCT CITATIONS

1. Dharan, A., et al. 2016. KIF5B and Nup358 cooperatively mediate the nuclear import of HIV-1 during infection. *PLoS Pathog.* 12: e1005700.

RESEARCH USE

For research use only, not for use in diagnostic procedures.