

FIP1L1 siRNA (h): sc-89183

BACKGROUND

The component of the Cleavage and Polyadenylation Specificity Factor (CPSF) complex plays an important role in the 3'-end formation of pre-mRNA. This complex recognizes the AAUAAA signal sequence and interacts with poly(A) polymerase to process and add to the poly(A) tail. FIP1L1 (FIP1-like 1), also known as pre-mRNA 3'-end-processing factor FIP1, FIP1 (factor interacting with PAP) and RHE (rearranged in hypereosinophilia), is a 594 amino acid nuclear protein that is a component of the CPSF complex. Within the complex, FIP1L1 contributes to the poly(A) recognition and stimulates poly(A) addition. Fusion of the genes encoding FIP1L1 and PDGFRA due to an interstitial deletion on chromosome 4q12 is the cause of hypereosinophilia syndrome, a rare blood disorder characterized by continuous overproduction of eosinophils in the bone marrow that leads to tissue infiltration and organ damage. There are three isoforms of FIP1L1 that are produced as a result of alternative splicing events.

REFERENCES

1. Preker, P.J., et al. 1995. The FIP1 gene encodes a component of a yeast pre-mRNA polyadenylation factor that directly interacts with poly(A) polymerase. *Cell* 81: 379-389.
2. Cools, J., et al. 2003. A tyrosine kinase created by fusion of the PDGFRA and FIP1L1 genes as a therapeutic target of imatinib in idiopathic hypereosinophilic syndrome. *N. Engl. J. Med.* 348: 1201-1214.
3. Griffin, J.H., et al. 2003. Discovery of a fusion kinase in EOL-1 cells and idiopathic hypereosinophilic syndrome. *Proc. Natl. Acad. Sci. USA* 100: 7830-7835.
4. Kaufmann, I., et al. 2004. Human Fip1 is a subunit of CPSF that binds to U-rich RNA elements and stimulates poly(A) polymerase. *EMBO J.* 23: 616-626.
5. Beausoleil, S.A., et al. 2004. Large-scale characterization of HeLa cell nuclear phosphoproteins. *Proc. Natl. Acad. Sci. USA* 101: 12130-12135.
6. Zhao, X., et al. 2005. A 57-nucleotide upstream early polyadenylation element in human papillomavirus type 16 interacts with hFip1, CstF-64, hnRNP C1/C2, and polypyrimidine tract binding protein. *J. Virol.* 79: 4270-4288.

CHROMOSOMAL LOCATION

Genetic locus: FIP1L1 (human) mapping to 4q12.

PRODUCT

FIP1L1 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 FIP1L1 shRNA Plasmid (h): sc-89183-SH and FIP1L1 shRNA (h) Lentiviral Particles: sc-89183-V as alternate gene silencing products.

For independent verification of FIP1L1 (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-89183A, sc-89183B and sc-89183C.

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

FIP1L1 siRNA (h) is recommended for the inhibition of FIP1L1 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

FIP1L1 (C-10): sc-398392 is recommended as a control antibody for monitoring of FIP1L1 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 FIP1L1 gene expression knockdown using RT-PCR Primer: FIP1L1 (h)-PR: sc-89183-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.