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Paf1 siRNA (m): sc-76035

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The Power to Question

BACKGROUND

In *Saccharomyces cerevisiae,* RNA polymerase II (RNAP II) mediates transcription elongation, and forms at least two distinct complexes. The first complex contains the Srb/mediator proteins, whereas the second complex, designated the Paf1 complex, contains Paf1, Cdc73, Hpr1, Ccr4, Rtf1, and Leo1. The Paf1 complex is required for full expression of a subset of yeast genes, particularly those responsive to signals from the Pkc1/MAP kinase cascade. The Paf1 complex mediates transcription elongation by physically associating with other transcription elongation factor complexes, including Spt16/Pob3 and Spt4/Spt5. It also plays an important role in the same regulatory pathways as Swi4/Swi6 and Mbp1/Swi6. Deletion of Paf1 or Cdc73 leads to increased recombination between direct repeats, while Paf1 and Ccr4 mutations demonstrate sensitivity to cell wall-damaging agents. Mutation of Rtf1 suppresses mutations in TBP, alters transcriptional start sites, and affects elongation.

REFERENCES

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3. Porter, S.E., Washburn, T.M., Chang, M. and Jaehning, J.A. 2002. The yeast pafl-rNA polymerase II complex is required for full expression of a subset of cell cycle-regulated genes. Eukaryot. Cell 1: 830-842.
4. Betz, J.L., Chang, M., Washburn, T.M., Porter, S.E., Mueller, C.L. and Jaehning, J.A. 2002. Phenotypic analysis of Paf1/RNA polymerase II complex mutations reveals connections to cell cycle regulation, protein synthesis, and lipid and nucleic acid metabolism. Mol. Genet. Genomics 268: 272-285.
5. Squazzo, S.L., Costa, P.J., Lindstrom, D.L., Kumer, K.E., Simic, R., Jennings, J.L., Link, A.J., Arndt, K.M. and Hartzog, G.A. 2002. The Paf1 complex physically and functionally associates with transcription elongation factors *in vivo*. EMBO J. 21: 1764-1774.

CHROMOSOMAL LOCATION

Genetic locus: Paf1 (mouse) mapping to 7 A3.

PRODUCT

Paf1 siRNA (m) 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 µM solution once resuspended using protocol below. Suitable for 50-100 transfections. Also see Paf1 shRNA Plasmid (m): sc-76035-SH and Paf1 shRNA (m) Lentiviral Particles: sc-76035-V as alternate gene silencing products.

For independent verification of Paf1 (m) gene silencing results, we also provide the individual siRNA duplex components. Each is available as 3.3 nmol of lyophilized siRNA. These include: sc-76035A, sc-76035B and sc-76035C.

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 µl of the RNAse-free water provided. Resuspension of the siRNA duplex in 330 µl of RNAse-free water makes a 10 µM solution in a 10 µM Tris-HCl, pH 8.0, 20 mM NaCl, 1 mM EDTA buffered solution.

APPLICATIONS

Paf1 siRNA (m) is recommended for the inhibition of Paf1 expression in mouse 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 µM in 66 µl. 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

Paf1 (E-7): sc-514491 is recommended as a control antibody for monitoring of Paf1 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).

RT-PCR REAGENTS

Semi-quantitative RT-PCR may be performed to monitor Paf1 gene expression knockdown using RT-PCR Primer: Paf1 (m)-PR: sc-76035-PR (20 µl). 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.