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Spt5 (yN-20): sc-26355

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The Power to Question

BACKGROUND

RNA polymerase II (RNAP II) comprises a 12 subunit protein complex that mediates transcription. RNAP II contains a unique carboxy terminal domain (CTD), which consists of 52 repeats of the consensus heptapeptide Tyr-Ser-Pro-Thr-Ser-Pro-Ser. The RNAP II isoform with an unphosphorylated CTD associates with transcription initiation complexes, whereas the isoform with a phosphorylated CTD is involved in transcription elongation. The suppressor of Ty proteins Spt4 and Spt5, which are highly conserved from yeast to human, form a complex designated DSIF (for DRB-sensitivity-inducing factor) with the phosphorylated form of RNAP II. The DSIF complex mediates interactions between RNAP II and nucleosomes to positively regulate transcription elongation in conjunction with the Paf1- and FACT-RNAP II associated complexes. The activity of Spt4 and Spt5 depends upon the length of the CTD of RNAP II, the presence of certain phosphoreceptors within the CTD, and the function of at least three CTD kinases. The Spt5 protein, which localizes to the nucleus, contains a highly acidic amino terminus and a novel six amino acid repeat (S-T/A-W-G-G-A/Q) in the carboxy terminus.

REFERENCES

1. Swanson, M.S., et al. 1991. SPT5, an essential gene important for normal transcription in *Saccharomyces cerevisiae*, encodes an acidic nuclear protein with a carboxy-terminal repeat. Mol. Cell. Biol. 11: 3009-3019.
2. Hartzog, G.A., et al. 1996. Identification and analysis of a functional human homolog of the SPT4 gene of *Saccharomyces cerevisiae*. Mol. Cell. Biol. 16: 2848-2856.
3. Hartzog, G.A., et al. 1998. Evidence that Spt4, Spt5, and Spt6 control transcription elongation by RNA polymerase II in *Saccharomyces cerevisiae*. Genes Dev. 12: 357-369.
4. Lindstrom, D.L. and Hartzog, G.A. 2001. Genetic interactions of Spt4-Spt5 and TFIIS with the RNA polymerase II CTD and CTD modifying enzymes in *Saccharomyces cerevisiae*. Genetics 159: 487-497.
5. Squazzo, S.L., et al. 2002. The Paf1 complex physically and functionally associates with transcription elongation factors *in vivo*. EMBO J. 21: 1764-1774.
6. Mandal, S.S., et al. 2002. FCP1, a phosphatase specific for the heptapeptide repeat of the largest subunit of RNA polymerase II, stimulates transcription elongation. Mol. Cell. Biol. 22: 7543-7552.
7. Kamada, K., et al. 2003. Molecular mechanism of recruitment of TFIIF- associating RNA polymerase C-terminal domain phosphatase (FCP1) by transcription factor IIF. Proc. Natl. Acad. Sci. USA 100: 2296-2299.
8. Friedl, E.M., et al. 2003. The C-terminal domain phosphatase and transcription elongation activities of FCP1 are regulated by phosphorylation. Proc. Natl. Acad. Sci. USA 100: 2328-2333.

SOURCE

Spt5 (yN-20) is an affinity purified goat polyclonal antibody raised against a peptide mapping near the N-terminus of Spt5 of *Saccharomyces cerevisiae* origin.

PRODUCT

Each vial contains 200 µg IgG in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin. Also available as TransCruz reagent for Gel Supershift and ChIP applications, sc-26355 X, 200 µg/0.1 ml.

Blocking peptide available for competition studies, sc-515648 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

APPLICATIONS

Spt5 (yN-20) is recommended for detection of Spt5 of *Saccharomyces cerevisiae* origin by Western Blotting (starting dilution 1:200, dilution range 1:100-1:1000) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

Spt5 (yN-20) X TransCruz antibody is recommended for Gel Supershift and ChIP applications.

Molecular Weight of Spt5: 160 kDa.

RECOMMENDED SECONDARY REAGENTS

To ensure optimal results, the following support (secondary) reagents are recommended: 1) Western Blotting: use donkey anti-goat IgG-HRP: sc-2020 (dilution range: 1:2000-1:100,000) or Cruz Marker™ compatible donkey anti-goat IgG-HRP: sc-2033 (dilution range: 1:2000-1:5000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto A Blocking Reagent: sc-2333 and Western Blotting Luminol Reagent: sc-2048.

SELECT PRODUCT CITATIONS

1. García, A., et al. 2012. Sub1 associates with Spt5 and influences RNA polymerase II transcription elongation rate. Mol. Biol. Cell 23: 4297-4312.
2. Mayer, A., et al. 2012. The spt5 C-terminal region recruits yeast 3' RNA cleavage factor I. Mol. Cell. Biol. 32: 1321-1331.

STORAGE

Store at 4° C, **DO NOT FREEZE**. Stable for one year from the date of shipment. Non-hazardous. No MSDS required.

RESEARCH USE

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

PROTOCOLS

See our web site at www.scbt.com or our catalog for detailed protocols and support products.