

SPT5 (E-8): sc-137231

BACKGROUND

SPT4 (also designated suppressor of Ty4 and p14) and SPT5 (also designated DSIF p160) are highly conserved proteins from yeast to humans. Nuclear SPT4 and SPT5 are involved in both DRB (5,6-dichloro-1- β -D-ribofuranosylbenzimidazole)-mediated transcriptional inhibition as well as the activation of transcriptional elongation by the HIV-1 protein Tat. SPT4 binds SPT5 to form the DSIF (DRB-sensitivity-inducing factor) complex, which binds RNA polymerase II and directly regulates elongation. SPT5 protein in mitotic HeLa cells migrates more slowly on SDS-PAGE than does SPT5 isolated from interphase cells, as a result of enhanced SPT5 phosphorylation. The C-terminal CTR1 domain of SPT5 is the substrate for P-TEFb phosphorylation, which is critical for SPT5 function as a regulator of transcriptional elongation.

REFERENCES

- Chiang, P.W., et al. 1996. Isolation and characterization of the human and mouse homologues (SUPT4H and Supt4h) of the yeast SPT4 gene. *Genomics* 34: 368-375.
- 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.
- Wada, T., et al. 1998. Evidence that P-TEFb alleviates the negative effect of DSIF on RNA polymerase II-dependent transcription *in vitro*. *EMBO J.* 17: 7395-7403.
- Wada, T., et al. 1998. DSIF, a novel transcription elongation factor that regulates RNA polymerase II processivity, is composed of human SPT4 and SPT5 homologs. *Genes Dev.* 12: 343-356.
- Yamaguchi, Y., et al. 1999. Structure and function of the human transcription elongation factor DSIF. *J. Biol. Chem.* 274: 8085-8092.
- Ivanov, D., et al. 2000. Domains in the SPT5 protein that modulate its transcriptional regulatory properties. *Mol. Cell. Biol.* 20: 2970-2983.

CHROMOSOMAL LOCATION

Genetic locus: SUPT5H (human) mapping to 19q13.2; Supt5 (mouse) mapping to 7 A3.

SOURCE

SPT5 (E-8) is a mouse monoclonal antibody raised against amino acids 61-360 mapping near the N-terminus of SPT5 of human origin.

PRODUCT

Each vial contains 200 μ g IgG₁ kappa light chain 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-137231 X, 200 μ g/0.1 ml.

STORAGE

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

APPLICATIONS

SPT5 (E-8) is recommended for detection of SPT5 of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 μ g per 100-500 μ g of total protein (1 ml of cell lysate)], immunofluorescence (starting dilution 1:50, dilution range 1:50-1:500) and solid phase ELISA (starting dilution 1:30, dilution range 1:30-1:3000).

SPT5 (E-8) is also recommended for detection of SPT5 in additional species, including equine, canine and porcine.

Suitable for use as control antibody for SPT5 siRNA (h): sc-38440, SPT5 siRNA (m): sc-38441, SPT5 shRNA Plasmid (h): sc-38440-SH, SPT5 shRNA Plasmid (m): sc-38441-SH, SPT5 shRNA (h) Lentiviral Particles: sc-38440-V and SPT5 shRNA (m) Lentiviral Particles: sc-38441-V.

SPT5 (E-8) X TransCruz antibody is recommended for Gel Supershift and ChIP applications.

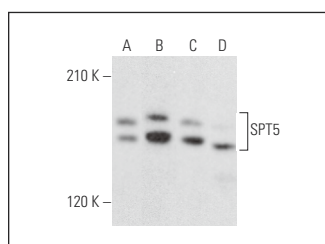
Molecular Weight of SPT5: 160 kDa.

Positive Controls: CCRF-CEM cell lysate: sc-2225, rat testis extract: sc-2400 or HeLa nuclear extract: sc-2120.

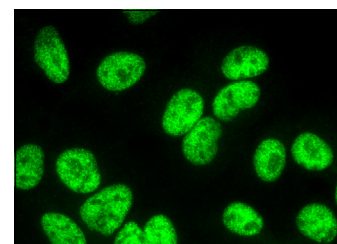
RECOMMENDED SUPPORT REAGENTS

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) Immunoprecipitation: use Protein A/G PLUS-Agarose: sc-2003 (0.5 ml agarose/2.0 ml). 3) 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.

DATA



SPT5 (E-8): sc-137231. Western blot analysis of SPT5 expression in CCRF-CEM (A) and OVCAR-3 (B) whole cell lysates, HeLa nuclear extract (C) and rat testis tissue extract (D).



SPT5 (E-8) sc-137231. Immunofluorescence staining of formalin-fixed Hep G2 cells showing nuclear localization.

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.