SANTA CRUZ BIOTECHNOLOGY, INC.

p-B-Myb (H-3): sc-377500

SANTA CRUZ BIOTECHNOLOGY

The Power to Question

BACKGROUND

A member of the myb proto-oncogene family, B-Myb is a cell cycle-regulated transcription factor that is essential for the transition from G_1 to S phase. This nuclear protein becomes phosphorylated at the onset of S phase by the cyclin A/Cdk2 complex. Ten phosphorylation sites have been identified and all sites were on either serine or threonine residues that were followed by a proline residue, suggesting that phosphorylation is due to a proline-directed kinase. Transactivation properties of B-Myb are apparently dependent upon the protein's hyperphosphorylation. Several phosphorylation sites, including threonine 447, threonine 490, threonine 497 and serine 581, are located near the protein's C-terminus. Phosphorylation of these C-terminal residues plays a critical role in enhancing the transcriptional activity of B-Myb. Poly(ADP-ribose) polymerase (PARP), which has a role in cellular proliferation, binds to B-Myb and thus enhances B-Myb transactivation. PARP is a B-Myb co-factor and promotes phosphorylation of B-Myb by the cyclin/Cdk2 complex.

REFERENCES

1. Johnson, T.K., et al. 1999. Phosphorylation of B-Myb regulates its transactivation potential and DNA binding. J. Biol. Chem. 274: 36741-36749.
2. Bartsch, O., et al. 1999. Identification of cyclin A/Cdk2 phosphorylation sites in B-Myb. Eur. J. Biochem. 260: 384-391.

CHROMOSOMAL LOCATION

Genetic locus: MYBL2 (human) mapping to 20q13.12; Mybl2 (mouse) mapping to 2 H2.

SOURCE

p-B-Myb (H-3) is a mouse monoclonal antibody epitope corresponding to a short amino acid sequence containing Thr 490 phosphorylated B-Myb of mouse origin.

PRODUCT

Each vial contains 200 µg IgG_{2b} kappa light chain in 1.0 ml of PBS with < 0.1% sodium azide and 0.1% gelatin.

p-B-Myb (H-3) is available conjugated to agarose (sc-377500 AC), 500 µg/ 0.25 ml agarose in 1 ml, for IP; to HRP (sc-377500 HRP), 200 µg/ml, for WB, IHC(P) and ELISA; to either phycoerythrin (sc-377500 PE), fluorescein (sc-377500 FITC), Alexa Fluor® 488 (sc-377500 AF488), Alexa Fluor® 546 (sc-377500 AF546), Alexa Fluor® 594 (sc-377500 AF594) or Alexa Fluor® 647 (sc-377500 AF647), 200 µg/ml, for WB (RGB), IF, IHC(P) and FCM; and to either Alexa Fluor® 680 (sc-377500 AF680) or Alexa Fluor® 790 (sc-377500 AF790), 200 µg/ml, for Near-Infrared (NIR) WB, IF and FCM.

Blocking peptide available for competition studies, sc-377500 P, (100 µg peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

STORAGE

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

APPLICATIONS

p-B-Myb (H-3) is recommended for detection of Thr 490 phosphorylated B-Myb of mouse, rat and human origin by Western Blotting (starting dilution 1:100, dilution range 1:100-1:1000), immunoprecipitation [1-2 µg per 100-500 µg 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).

p-B-Myb (H-3) is also recommended for detection of correspondingly phosphorylated B-Myb in additional species, including equine, canine, bovine, porcine and avian.

Suitable for use as control antibody for B-Myb siRNA (h): sc-43523, B-Myb siRNA (m): sc-43524, B-Myb siRNA (r): sc-108008, B-Myb shRNA Plasmid (h): sc-43523-SH, B-Myb shRNA Plasmid (m): sc-43524-SH, B-Myb shRNA Plasmid (r): sc-108008-SH, B-Myb shRNA (h) Lentiviral Particles: sc-43523-V, B-Myb shRNA (m) Lentiviral Particles: sc-43524-V and B-Myb shRNA (r) Lentiviral Particles: sc-108008-V.

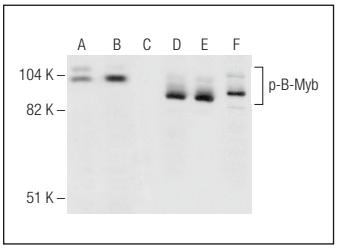
Molecular Weight of p-B-Myb: 110 kDa.

Positive Controls: HEK293 whole cell lysate: sc-45136 or Raji whole cell lysate: sc-364236.

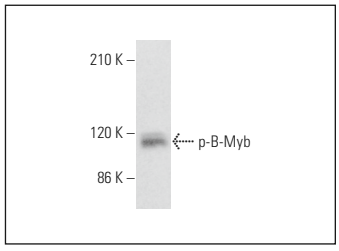
RECOMMENDED SUPPORT REAGENTS

To ensure optimal results, the following support reagents are recommended: 1) Western Blotting: use m-IgGκ BP-HRP: sc-516102 or m-IgGκ BP-HRP (Cruz Marker): sc-516102-CM (dilution range: 1:1000-1:10000), Cruz Marker™ Molecular Weight Standards: sc-2035, TBS Blotto B Blocking Reagent: sc-2335 (use 50 mM NaF, sc-24988, as diluent), Lambda Phosphatase: sc-200312A 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-IgGκ BP-FITC: sc-516140 or m-IgGκ 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



Western blot analysis of B-Myb phosphorylation in untreated (**A**,**D**), PMA treated (**B**,**E**) and PMA and lambda protein phosphatase (sc-200312A) treated (**C**,**F**) K-562 nuclear extracts. Antibodies tested include p-B-Myb (H-3): sc-377500 (**A**,**B**,**C**) and B-Myb (C-20): sc-725 (**D**,**E**,**F**).



p-B-Myb (H-3): sc-377500. Western blot analysis of B-Myb phosphorylation in Raji whole cell lysate.

RESEARCH USE

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