SANTA CRUZ BIOTECHNOLOGY, INC.

p-Rb (B-8): sc-377541



BACKGROUND

Pediatric cancer retinoblastoma and the formation of other human tumors can be attributed to mutations in the retinoblastoma tumor suppressor gene (Rb). The Rb protein regulates differentiation, apoptosis, and cell cycle control by coordinating the cell cycle at G₁-S with transcriptional machinery. During G₁, cyclin D-dependent kinase-mediated phosphorylation of Rb at Ser 795 marks the conversion of Rb from a transcriptionally repressive, hypophosphorylated state to an inactive, phosphorylated state, which may be sustained through mitosis by differential phosphorylation of up to 16 putative serine or threonine residues, including Ser 249/Thr 252, Thr 373, Thr 356, Ser 780, Ser 807/ Ser 811 and Thr 821/Thr 826. Hypophosphorylated Rb represses the transcription of genes controlling the cell cycle through direct protein-protein interactions and through the recruitment of histone deacetylase.

CHROMOSOMAL LOCATION

Genetic locus: RB1 (human) mapping to 13q14.2; Rb1 (mouse) mapping to 14 D3.

SOURCE

p-Rb (B-8) is a mouse monoclonal antibody raised against a short amino acid sequence containing Thr 356 phosphorylated Rb of human origin.

PRODUCT

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

Blocking peptide available for competition studies, sc-377541 P, (100 μ g peptide in 0.5 ml PBS containing < 0.1% sodium azide and 0.2% stabilizer protein).

APPLICATIONS

p-Rb (B-8) is recommended for detection of Thr 356 phosphorylated Rb 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), immunohistochemistry (including paraffinembedded sections) (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-Rb (B-8) is also recommended for detection of correspondingly phosphorylated Rb in additional species, including equine, bovine and porcine.

Suitable for use as control antibody for Rb siRNA (h): sc-29468, Rb siRNA (m): sc-29469, Rb shRNA Plasmid (h): sc-29468-SH, Rb shRNA Plasmid (m): sc-29469-SH, Rb shRNA (h) Lentiviral Particles: sc-29468-V and Rb shRNA (m) Lentiviral Particles: sc-29469-V.

Molecular Weight (predicted) of p-Rb: 106 kDa.

Molecular Weight (observed) of p-Rb: 107-140 kDa.

Positive Controls: Rb (h): 293T Lysate: sc-114014, SK-LMS-1 cell lysate: sc-3813 or MOLT-4 cell lysate: sc-2233.

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 MarkerTM 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-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. 4) Immunohistochemistry: use m-IgG κ BP-HRP: sc-516102 with DAB, 50X: sc-24982 and Immunohistomount: sc-45086, or Organo/Limonene Mount: sc-45087.

DATA





Western blot analysis of Rb phosphorylation in nontransfected: sc-117752 (**A**,**D**), untreated human Rb transfected: sc-114014 (**B**,**E**) and lambda protein phosphatase (sc-200312A) treated human Rb transfected: sc-114014 (**C**,**F**) 2937 whole cell lysates. Antibodies tested include p-Rb (B-8): sc-377541 (**A**,**B**,**C**) and Rb (M-153): sc-7305 (**D**,**E**,**F**).

p-Rb (B-8): sc-377541. Immunoperoxidase staining of formalin fixed, paraffin-embedded human duodenum tissue showing nuclear staining of glandular cells.

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 for detailed protocols and support products.