#8147 Store at -20C

Phospho-Rb (Ser608) (D10F2) Rabbit mAb

Cell Signaling TECHNOLOGY®

✓ 100 µl
(10 western blots)

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New 12/11

This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Entrez-Gene ID #5925
Swiss-Prot Acc. #P06400

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP	H, (Mk)	110 kDa	Rabbit IgG**
Endogenous			

Storage: Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at –20°C. *Do not aliquot the antibody.*

***Species cross-reactivity is determined by western blot.**

****Anti-rabbit secondary antibodies must be used to detect this antibody.**

Recommended Antibody Dilutions:

Western blotting	1:1000
Immunoprecipitation	1:50

For application specific protocols please see the web page for this product at www.cellsignal.com.

Please visit www.cellsignal.com for a complete listing of recommended companion products.

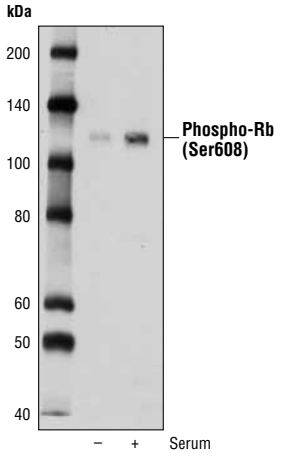
Background: The retinoblastoma tumor suppressor protein, Rb, regulates cell proliferation by controlling progression through the restriction point within the G1-phase of the cell cycle (1). Rb has three functionally distinct binding domains and interacts with critical regulatory proteins including the E2F family of transcription factors, c-Abl tyrosine kinase, and proteins with a conserved LXCXE motif (2-4). Cell cycle-dependent phosphorylation by a CDK inhibits Rb target binding and allows cell cycle progression (5). Rb inactivation and subsequent cell cycle progression likely requires an initial phosphorylation by cyclin D-CDK4/6 followed by cyclin E-CDK2 phosphorylation (6). Specificity of different CDK/cyclin complexes has been observed *in vitro* (6-8) and cyclin D1 is required for Ser780 phosphorylation *in vivo* (9).

Specificity/Sensitivity: Phospho-Rb (Ser608) (D10F2) Rabbit mAb recognizes endogenous levels of Rb protein only when phosphorylated at Ser608.

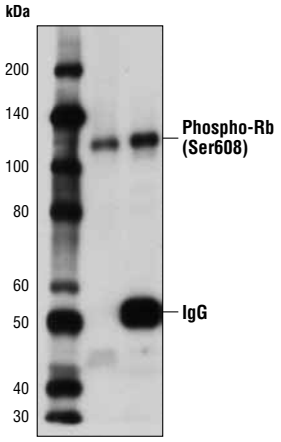
Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser608 of human Rb protein.

Background References:

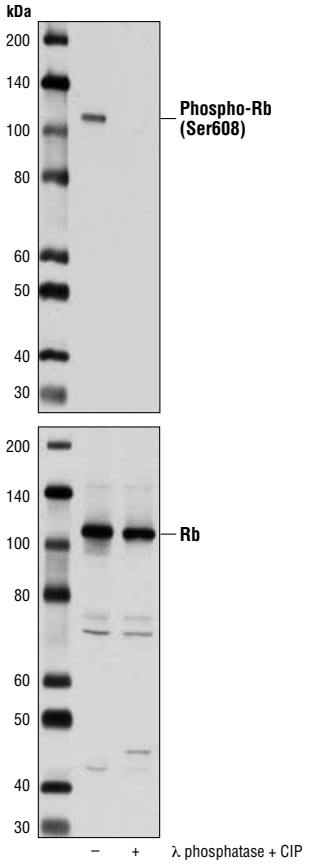
(1) Sherr, C.J. (1996) *Science* 274, 1672-7.
(2) Nevins, J.R. (1992) *Science* 258, 424-9.
(3) Welch, P.J. and Wang, J.Y. (1993) *Cell* 75, 779-90.
(4) Hu, Q.J. et al. (1990) *EMBO J* 9, 1147-55.
(5) Knudsen, E.S. and Wang, J.Y. (1997) *Mol Cell Biol* 17, 5771-83.
(6) Lundberg, A.S. and Weinberg, R.A. (1998) *Mol Cell Biol* 18, 753-61.
(7) Connell-Crowley, L. et al. (1997) *Mol Biol Cell* 8, 287-301.
(8) Kitagawa, M. et al. (1996) *EMBO J* 15, 7060-9.
(9) Geng, Y. et al. (2001) *Proc Natl Acad Sci USA* 98, 194-9.



Western blot analysis of extracts from WI-38 cells, serum-starved for 3 days (-) or serum-starved for 3 days followed by treatment with 10% serum for 2 days (+), using Phospho-Rb (Ser608) (D10F2) Rabbit mAb.



Immunoprecipitation of phospho-Rb (Ser608) from WI-38 cell extracts using Phospho-Rb (Ser608) (D10F2) Rabbit mAb (lane 2). Western blot was performed using the same antibody. Lane 1 is 10% input.



Western blot analysis of extracts from MCF7 cells, untreated (-) or treated with calf intestinal phosphatase (CIP) and λ phosphatase (+), using Phospho-Rb (Ser608) (D10F2) Rabbit mAb (upper) or Rb (D20) Rabbit mAb #9313 (lower).

IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.

Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide
Species Cross-Reactivity Key: **H**—human **M**—mouse **R**—rat **Hm**—hamster **Mk**—monkey **Mi**—mink **C**—chicken **Dm**—D. melanogaster **X**—Xenopus **Z**—zebrafish **B**—bovine **Dg**—dog **Pg**—pig **Sc**—S. cerevisiae **Ce**—C. elegans **Hr**—horse **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.

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