Store at 4°C

#8880

# Phospho-Rb (Ser807/811) (D20B12) XP® Rabbit mAb (Alexa Fluor® 594 Conjugate)

Cell Signaling TECHNOLOGY®

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**For Research Use Only. Not For Use In Diagnostic Procedures.**

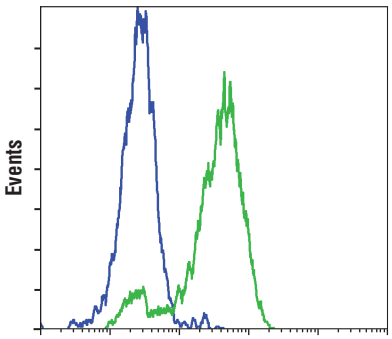
**Entrez-Gene ID** #5925
**UniProt Acc.** #P06400

| Applications | Species Cross-Reactivity* | Isotype |
|---|---|---|
| IF-IC, F | H, M, R, Mk | Rabbit IgG |
| Endogenous | | |

**Description:** This Cell Signaling Technology antibody is conjugated to Alexa Fluor® 549 fluorescent dye and tested in-house for direct flow cytometry and immunofluorescent analysis in human cells. The antibody is expected to exhibit the same species cross-reactivity as the unconjugated Phospho-Rb (Ser807/811) (D20B12) XP® Rabbit mAb #8516.

**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 (Ser807/811) (D20B12) XP® Rabbit mAb (Alexa Fluor® 594 Conjugate) recognizes endogenous levels of Rb protein only when phosphorylated at Ser807, Ser811, or at both sites. This antibody does not cross-react with Rb phosphorylated at Ser608.




*Flow cytometic analysis of BT-549 (blue) and Jurkat (green) cells using Phospho-Rb (Ser807/811) (D20B12) XP® Rabbit mAb (Alexa Fluor® 594 Conjugate).*

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser807/811 of human Rb protein.

**Storage:** Supplied in PBS (pH 7.2), less than 0.1% sodium azide and 2 mg/ml BSA. Store at 4°C. *Protect from light. Do not freeze.*

***Species cross-reactivity other than human is determined by western using the unconjugated antibody.**

**Recommended Antibody Dilutions:**

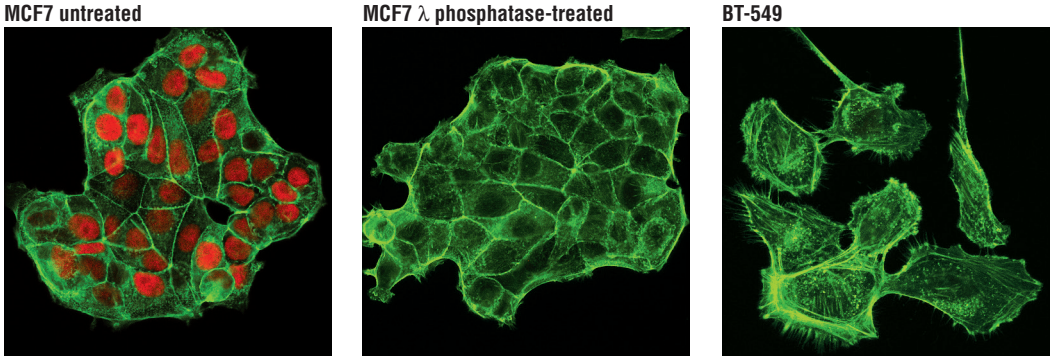
| | |
|---|---|
| Flow Cytometry | 1:50 |
| Immunofluorescence (IF-IC) | 1:400 |

**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 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.




*Confocal immunofluorescent analysis of MCF7 cells, untreated (left) or λ phosphatase-treated (middle), and BT-549 cells (right), using Phospho-Rb (Ser807/811) (D20B12) XP® Rabbit mAb (Alexa Fluor® 594 Conjugate) (red). Actin filaments were labeled with Alexa Fluor® 488 phalloidin (green).*



**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 **All**—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.