#4513 Store at –20°C

PLK1 (208G4) Rabbit mAb

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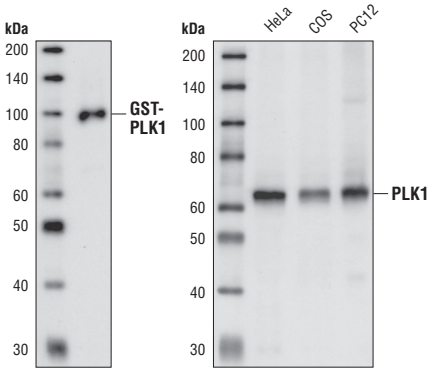
Entrez-Gene ID #5347
UniProt ID #P53350

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, IP, IHC-P Endogenous	H, R, Mk	62 kDa	Rabbit IgG**

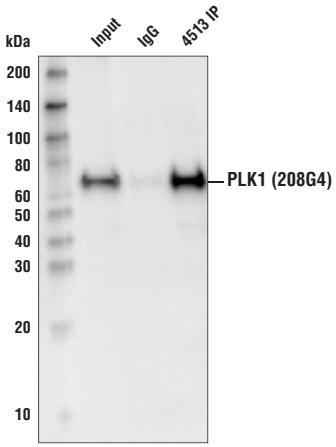
Background: At least 4 distinct polo-like kinases exist in mammalian cells: PLK1, PLK2, PLK3 and SAK (1). PLK1 apparently plays many roles during mitosis, particularly in regulating mitotic entry and exit. The mitosis promoting factor (MPF), cdc2/cyclin B1, is activated by dephosphorylation of cdc2 (Thr14/Tyr15) by cdc25C. PLK1 phosphorylates cdc25C at Ser198 and cyclin B1 at Ser133 causing translocation of these proteins from the cytoplasm to the nucleus (2–5). PLK1 phosphorylation of Myt1 at Ser426 and Thr495 has been proposed to inactivate Myt1, one of the kinases known to phosphorylate cdc2 at Thr14/Tyr15 (6). Polo-like kinases also phosphorylate the cohesin subunit SCC1, causing cohesin displacement from chromosome arms that allow for proper cohesin localization to centromeres (7). Mitotic exit requires activation of the anaphase promoting complex (APC) (8), a ubiquitin ligase responsible for removal of cohesin at centromeres, and degradation of securin, cyclin A, cyclin B1, Aurora A and cdc20 (9). PLK1 phosphorylation of the APC subunits Apc1, cdc16, and cdc27 has been demonstrated *in vitro* and has been proposed as a mechanism by which mitotic exit is regulated (10,11).

Specificity/Sensitivity: PLK1 (208G4) Rabbit mAb detects endogenous levels of of total PLK1 protein.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding Pro339 of human PLK1.



Western blot analysis of GST-PLK1 fusion protein and extracts from HeLa, COS and PC12 cells, using PLK1 (208G4) Rabbit mAb.



Immunoprecipitation/western blot analysis of lysates from HeLa cells. Lane 1 contains lysate input (10%), lane 2 was immunoprecipitated with non-specific rabbit IgG, lane 3 was immunoprecipitated with PLK1 (208G4) Rabbit mAb #4513. Western blot analysis was performed using PLK1 (208G4) Rabbit mAb #4513 along with a Mouse anti-rabbit IgG (confirmation specific) (L27A9) mAb (HRP conjugate)#5127 secondary.

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	1:100
Immunohistochemistry (Paraffin)	1:50†
Unmasking buffer:	Citrate
Antibody diluent:	SignalStain® Antibody Diluent #8112
Detection reagent:	SignalStain® Boost (HRP, Rabbit) #8114

†*Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.*

For product 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.

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.

U.S. Patent No. 5,675,063

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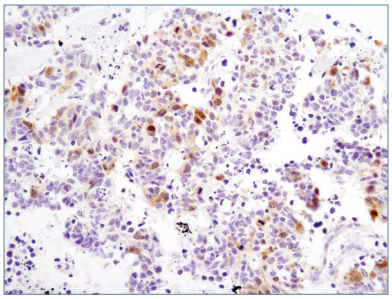
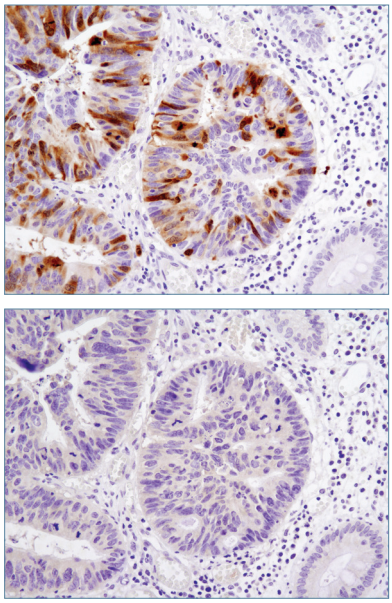
Applications Key: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide

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.



Immunohistochemical analysis of paraffin-embedded human lung carcinoma using PLK1 (208G4) Rabbit mAb.



Immunohistochemical analysis of paraffin-embedded human colon carcinoma using PLK1 (208G4) Rabbit mAb in the presence of control peptide (upper) or antigen-specific peptide (lower).

Background References:

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(5) Jackman, M. et al. (2003) *Nat. Cell Biol.* 5, 143–148.

(6) Nakajima, H. et al. (2003) *J. Biol. Chem.* 278, 25277–25280.

(7) Sumara, I. et al. (2002) *Mol. Cell* 9, 515–525.

(8) Hauf, S. et al. (2001) *Science* 293, 1320–1323.

(9) Peters, J.M. (1999) *Exp. Cell Res.* 248, 339–349.

(10) Kraft, C. et al. (2003) *EMBO J.* 22, 6598–6609.

(11) Kotani, S. et al. (1998) *Mol. Cell* 1, 371–380.