Revision 1

Phospho-Chk1 (Ser280) Antibody

#2347 Store at -20C

Cell Signaling TECHNOLOGY®

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Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
W	H	Endogenous	56	Rabbit	#O14757	1111

Product Usage Information

Application	Dilution
Western Blotting	1:1000

Storage

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

Specificity/Sensitivity

Phospho-Chk1 (Ser280) Antibody detects endogenous levels of Chk1 only when phosphorylated at serine 280.

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser280 of human Chk1. Antibodies are purified by protein A and peptide affinity chromatography.

Background

Chk1 kinase acts downstream of ATM/ATR kinase and plays an important role in DNA damage checkpoint control, embryonic development, and tumor suppression (1). Activation of Chk1 involves phosphorylation at Ser317 and Ser345 by ATM/ATR, followed by autophosphorylation of Ser296. Activation occurs in response to blocked DNA replication and certain forms of genotoxic stress (2). While phosphorylation at Ser345 serves to localize Chk1 to the nucleus following checkpoint activation (3), phosphorylation at Ser317 along with site-specific phosphorylation of PTEN allows for re-entry into the cell cycle following stalled DNA replication (4). Chk1 exerts its checkpoint mechanism on the cell cycle, in part, by regulating the cdc25 family of phosphatases. Chk1 phosphorylation of cdc25A targets it for proteolysis and inhibits its activity through 14-3-3 binding (5). Activated Chk1 can inactivate cdc25C via phosphorylation at Ser216, blocking the activation of cdc2 and transition into mitosis (6). Centrosomal Chk1 has been shown to phosphorylate cdc25B and inhibit its activation of CDK1-cyclin B1, thereby abrogating mitotic spindle formation and chromatin condensation (7). Furthermore, Chk1 plays a role in spindle checkpoint function through regulation of aurora B and BubR1 (8). Research studies have implicated Chk1 as a drug target for cancer therapy as its inhibition leads to cell death in many cancer cell lines (9).
Akt has been shown to phosphorylate Chk1 at Ser280 in vitro, and preliminary data suggests that Akt activity may prevent Chk1 activation (5).

Background References

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3. Jiang, K. et al. (2003) *J Biol Chem* 278, 25207-17.
4. Martin, S.A. and Ouchi, T. (2008) *Mol Cancer Ther* 7, 2509-16.
5. Chen, M.S. et al. (2003) *Mol Cell Biol* 23, 7488-97.
6. Zeng, Y. et al. (1998) *Nature* 395, 507-10.
7. Löffler, H. et al. (2006) *Cell Cycle* 5, 2543-7.
8. Zachos, G. et al. (2007) *Dev Cell* 12, 247-60.
9. Garber, K. (2005) *J Natl Cancer Inst* 97, 1026-8.
10. Shtivelman, E. et al. (2002) *Curr. Biol.* 12, 919-924.

Species Reactivity

Species reactivity is determined by testing in at least one approved application (e.g., western blot).

Western Blot Buffer

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

Applications Key

W: Western Blotting

Cross-Reactivity Key

H: Human

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