Revision 1

Phospho-Cyclin E1 (Thr62) Antibody

#4136 Store at -20C

Cell Signaling TECHNOLOGY®

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For Research Use Only. Not for Use in Diagnostic Procedures.

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:
W, IP, IHC-P, FC-FP	H	Endogenous	48	Rabbit

Product Usage Information

Application	Dilution
Western Blotting	1:1000
Immunoprecipitation	1:100
Immunohistochemistry (Paraffin)	1:100
Flow Cytometry (Fixed/Permeabilized)	1:50

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-Cyclin E1 (Thr62) Antibody detects endogenous levels of cyclin E only when phosphorylated at threonine 62 (cyclin E1 isoform 2) or threonine 77 (cyclin E1 isoform 1).

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Thr62 of human cyclin E1. Antibodies are purified by protein protein A and peptide affinity chromatography.

Background

Cyclin E1 and cyclin E2 can associate with and activate CDK2 (1). Upon DNA damage, upregulation/activation of the CDK inhibitors p21 Waf1/Cip1 and p27 Kip1 prevent cyclin E/CDK2 activation, resulting in G1/S arrest. When conditions are favorable for cell cycle progression, cyclin D/CDK4/6 phosphorylates Rb and is thought to reduce the activity of p21 Waf1/Cip1 and p27 Kip1, allowing subsequent activation of cyclin E/CDK2 (1,2). Cyclin E/CDK2 further phosphorylates Rb to allow progression into S-phase, where cyclin E/CDK2 is thought to phosphorylate and activate multiple proteins involved in DNA synthesis (2,3). Turnover of cyclin E is largely controlled by phosphorylation that results in SCFFbw7-mediated ubiquitination and proteasome-dependent degradation (4,5). Cyclin E1 is phosphorylated at multiple sites *in vivo* including Thr62, Ser88, Ser72, Thr380, and Ser384, and is controlled by at least two kinases, CDK2 and GSK-3 (6,7).

Background References

1. Lauper, N. et al. (1998) *Oncogene* 17, 2637-43.
2. Lundberg, A.S. and Weinberg, R.A. (1998) *Mol Cell Biol* 18, 753-61.
3. Ewen, M.E. (2000) *Genes Dev* 14, 2265-70.
4. Won, K.A. and Reed, S.I. (1996) *EMBO J* 15, 4182-93.
5. Koepp, D.M. et al. (2001) *Science* 294, 173-7.
6. Welcker, M. et al. (2003) *Mol Cell* 12, 381-92.
7. Ye, X. et al. (2004) *J Biol Chem* 279, 50110-9.

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 **IP:** Immunoprecipitation **IHC-P:** Immunohistochemistry (Paraffin) **FC-FP:** Flow Cytometry (Fixed/Permeabilized)

Cross-Reactivity Key

H: Human

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