

#2600 Store at -20°C

Phospho-HP1 γ (Ser83) Antibody


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

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source:	UniProt ID:	Entrez-Gene Id:
WB, IP, IF-IC	H M R Mk	Endogenous	22	Rabbit	#Q13185	11335

Product Usage Information

Application

Western Blotting
Immunoprecipitation
Immunofluorescence (Immunocytochemistry)

Dilution

1:1000
1:25
1:200

Storage

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

Specificity / Sensitivity

Phospho-HP1 γ (Ser83) Antibody detects endogenous levels of HP1 γ protein only when phosphorylated on Ser83 (also referred to as Ser93 of the unprocessed form of HP1 γ). This antibody does not cross-react with HP1 α or HP1 β proteins.

Species predicted to react based on 100% sequence homology:

D. melanogaster, Bovine, Horse

Source / Purification

Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to amino acids surrounding Ser83 of human HP1 γ . Antibodies are purified by affinity chromatography.

Background

Heterochromatin protein 1 (HP1) is a family of heterochromatic adaptor molecules involved in both gene silencing and higher order chromatin structure (1). All three HP1 family members (α , β , and γ) are primarily associated with centromeric heterochromatin; however, HP1 β and γ also localize to euchromatic sites in the genome (2,3). HP1 proteins are approximately 25 kDa in size and contain a conserved amino-terminal chromodomain, followed by a variable hinge region and a conserved carboxy-terminal chromoshadow domain. The chromodomain facilitates binding to histone H3 tri-methylated at Lys9, a histone "mark" closely associated with centromeric heterochromatin (4,5). The variable hinge region binds both RNA and DNA in a sequence-independent manner (6). The chromoshadow domain mediates the dimerization of HP1 proteins, in addition to binding multiple proteins implicated in gene silencing and heterochromatin formation, including the SUV39H histone methyltransferase, the DNMT1 and DNMT3a DNA methyltransferases, and the p150 subunit of chromatin-assembly factor-1 (CAF1) (7-9). In addition to contributing to heterochromatin formation and propagation, HP1 and SUV39H are also found complexed with retinoblastoma (Rb) and E2F6 proteins, both of which function to repress euchromatic gene transcription in quiescent cells (10,11). HP1 proteins are subject to multiple types of post-translational modifications, including phosphorylation, acetylation, methylation, ubiquitination, and sumoylation, suggesting multiple means of regulation (12-14).

HP1 γ is phosphorylated on Ser83 by protein kinase A (PKA) *in vitro*, and activation of PKA by forskolin and IBMX treatment leads to increased phosphorylation *in vivo* (14). Phosphorylation of HP1 γ on Ser83 also increases during mitosis as demonstrated by the Phospho-HP1 γ (Ser83) Antibody, which shows increased immunofluorescent staining in untreated mitotic cells and increased Western blot signal in lysates from cells arrested in mitosis by treatment with paclitaxel. Phosphorylation of Ser83 only occurs on a subpopulation of HP1 γ found associated with euchromatin, specifically HP1 γ bound to coding regions of active genes (14). This phosphorylation impairs the ability of HP1 γ to silence transcription and may be a marker for transcription elongation (14).

Background References

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6. Muchardt, C. et al. (2002) *EMBO Rep.* 3, 975-981.

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11. Ogawa, H. et al. (2002) *Science* 296, 1132-1136.
12. Minc, E. et al. (1999) *Chromosoma* 108, 220-234.
13. Zhao, T. et al. (2001) *J. Biol. Chem.* 276, 9512-9518.
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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

WB: Western Blotting **IP:** Immunoprecipitation **IF-IC:** Immunofluorescence (Immunocytochemistry)

Cross-Reactivity Key

H: human **M:** mouse **R:** rat **Hm:** hamster **Mk:** monkey **Vir:** virus **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
GP: Guinea Pig **Rab:** rabbit **All:** all species expected

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