

**#9701** Store at -20°C

# Phospho-Histone H3 (Ser10) Antibody

- Small 100 µl (10 western blots)
- Large 300 µl (30 western blots)



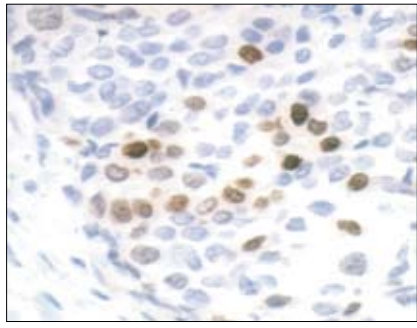
**Orders** ■ 877-616-CELL (2355)  
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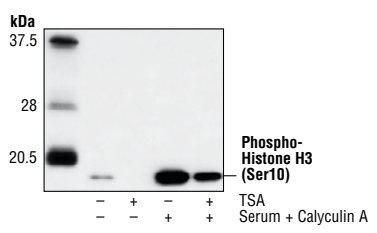
This product is intended for research purposes only. This product is not intended to be used for therapeutic or diagnostic purposes in humans or animals.

Applications	Species Cross-Reactivity*	Molecular Wt.	Source
W, IP, IHC-P, IHC-F, IF-IC, F Endogenous	H, M, R, Mk, Sc, C, Dm Z, (X)	17 kDa	Rabbit**

**Background:** Modulation of chromatin structure plays an important role in the regulation of transcription in eukaryotes. The nucleosome, made up of four core histone proteins (H2A, H2B, H3 and H4), is the primary building block of chromatin (1). The amino-terminal tails of core histones undergo various post-translational modifications, including acetylation, phosphorylation, methylation and ubiquitination (2-5). These modifications occur in response to various stimuli and have a direct effect on the accessibility of chromatin to transcription factors and, therefore, on gene expression (6). In most species, histone H2B is primarily acetylated at Lys5, 12, 15 and 20 (4,7). Histone H3 is primarily acetylated at Lys9, 14, 18 and 23 (2,3). Acetylation at Lys9 appears to have a dominant role in histone deposition and chromatin assembly in some organisms (2,3). Phosphorylation at Ser10, Ser28 and Thr11 of histone H3 is tightly correlated with chromosome condensation during both mitosis and meiosis (8-10). Phosphorylation of Thr3 of histone H3 is highly conserved among many species and is catalyzed by the kinase haspin. Immunostaining with phospho-specific antibodies in mammalian cells reveals mitotic phosphorylation of H3 Thr3 in prophase and its dephosphorylation during anaphase (11).



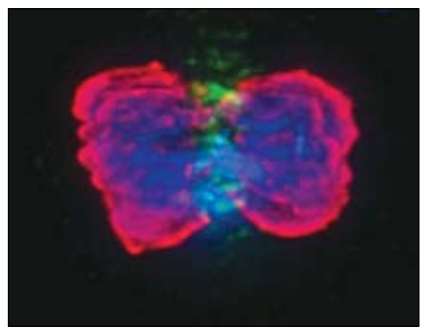
Immunohistochemical staining of phosphorylated histone H3 in paraffin-embedded human breast carcinoma showing nuclear localization, using Phospho-Histone H3 (Ser10) Antibody.



Western blot analysis of whole cell lysates of NIH/3T3 cells, untreated, treated with TSA (to induce histone acetylation), serum plus calyculin A (to induce phosphorylation of H3) or both, using Phospho-Histone H3 (Ser10) Antibody.

**Specificity/Sensitivity:** Phospho-Histone H3 (Ser10) Antibody detects endogenous levels of histone H3 only when phosphorylated at serine 10. The antibody does not cross-react with other phosphorylated histones or with acetylated histones.

**Source/Purification:** Polyclonal antibodies are produced by immunizing animals with a synthetic phosphopeptide corresponding to residues surrounding Ser10 of human histone H3. Antibodies are purified by protein A and peptide affinity chromatography.



Confocal microscopic image of a mitotic HeLa cell labeled with Phospho-Histone H3 (Ser10) Antibody (red) and Survivin (6E4) Mouse mAb #2802 (green). Blue pseudocolor = DRAQ5 #4084 (fluorescent DNA dye).

**Entrez-Gene ID** #8352  
**Swiss-Prot Acc.** #P68431

**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.

**\*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	1:50
Immunohistochemistry (Paraffin)	1:200†
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.	
Immunohistochemistry (Frozen)	1:400†
Fixative:	10% Neutral buffered formalin
Detection reagent:	SignalStain® Boost (HRP, Rabbit) #8114
† Optimal IHC dilutions determined using SignalStain® Boost IHC Detection Reagent.	
Immunofluorescence (IF-IC)	1:100
Flow Cytometry	1:50

For application specific protocols please see the web page for this product at [www.cellsignal.com](http://www.cellsignal.com).

Please visit [www.cellsignal.com](http://www.cellsignal.com) for a complete listing of recommended companion products.

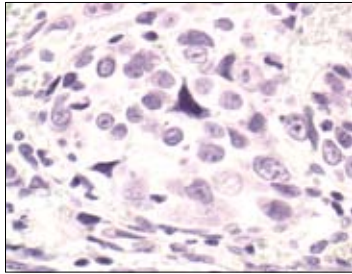
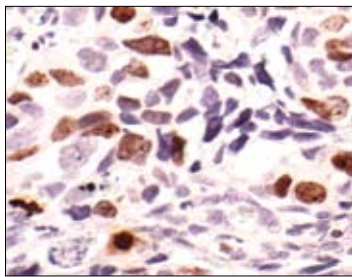
**Background References:**

- (1) Workman, J.L. and Kingston, R.E. (1998) *Annu. Rev. Biochem.* 67, 545-579.
- (2) Hansen, J.C. et al. (1998) *Biochemistry* 37, 17637-17641.
- (3) Strahl, B.D. and Allis, C.D. (2000) *Nature* 403, 41-45.
- (4) Cheung, P. et al. (2000) *Cell* 103, 263-271.
- (5) Bernstein, B.E. and Schreiber, S.L. (2002) *Chem. Biol.* 9, 1167-1173.
- (6) Jaskelioff, M. and Peterson, C.L. (2003) *Nat. Cell Biol.* 5, 395-399.
- (7) Thorne, A.W. et al. (1990) *Eur. J. Biochem.* 193, 701-713.
- (8) Hendzel, M.J. et al. (1997) *Chromosoma* 106, 348-360.
- (9) Goto, H. et al. (1999) *J. Biol. Chem.* 274, 25543-25549.
- (10) Preuss, U. et al. (2003) *Nucleic Acids Res.* 31, 878-885.
- (11) Dai, J. et al. (2005) *Genes Dev.* 19, 472-488

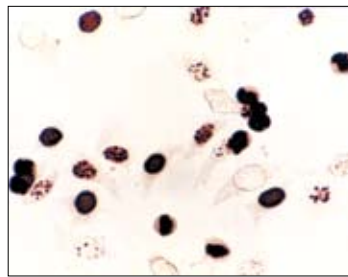
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**IMPORTANT: For western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.**

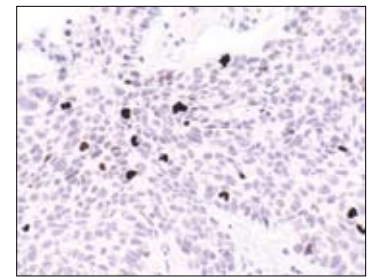
**Applications Key:** W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide  
**Species Cross-Reactivity Key:** H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish 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.



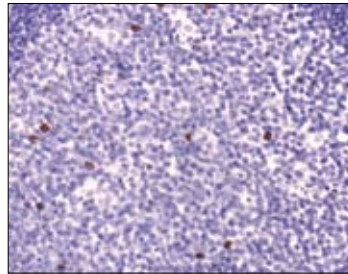
Immunohistochemical analysis of paraffin-embedded human colon carcinoma, untreated (upper) or  $\lambda$  phosphatase-treated (lower), using Phospho-Histone H3 (Ser10) Antibody.



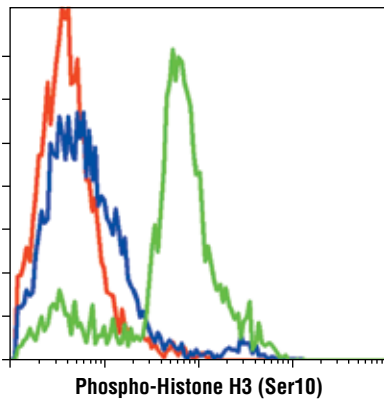
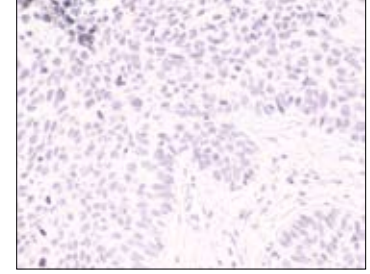
Immunocytochemical staining of NIH/3T3 cells, nocodazole-treated (2 ug/ml) and grown in 10% serum, showing cells undergoing mitosis, using Phospho-Histone (Ser10) Antibody.



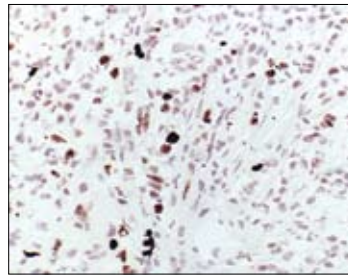
Immunohistochemical analysis of paraffin-embedded human lung carcinoma using Phospho-Histone H3 (Ser10) Antibody in the presence of control peptide (upper) or Phospho-Histone H3 (Ser10) Blocking Peptide #1000 (lower).



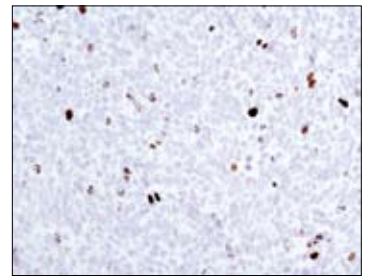
Immunohistochemical analysis of paraffin-embedded human tonsil using Phospho-Histone H3 (Ser10) Antibody.



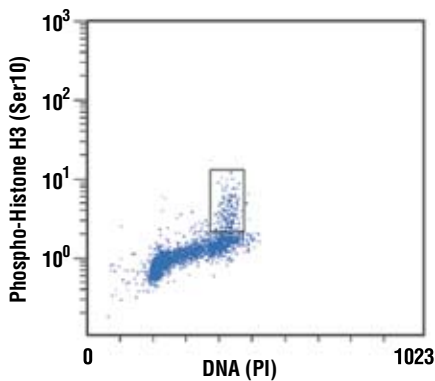
Flow cytometric analysis of Phospho-Histone H3 (Ser10) Antibody staining of untreated (blue) or serum/calyculin treated (green) Ramos cells compared to a nonspecific negative control antibody (red).



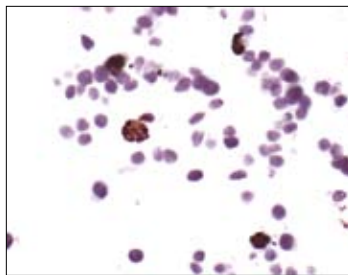
Immunohistochemical analysis of frozen H1650 xenograft showing staining of mitotic cells using Phospho-Histone H3 (Ser10) Antibody.



Immunohistochemical analysis of paraffin-embedded 4T1 syngeneic mouse tumor using Phospho-Histone H3 (Ser10) Antibody.



Flow cytometric analysis of untreated Jurkat cells, using Phospho-Histone H3 (Ser10) Antibody versus propidium iodide (DNA content). The box indicates phospho-histone H3 positive cells.



Immunohistochemical analysis of HT29 cells using Phospho-Histone H3 (Ser10) Antibody. Note the specific staining of mitotic cells.