#12230 Store at 4°C

# Histone H3 (D1H2) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate)

☑ 100 µl
(50 tests)

Cell Signaling TECHNOLOGY®

**Orders** ■ 877-616-CELL (2355)
orders@cellsignal.com
**Support** ■ 877-678-TECH (8324)
info@cellsignal.com
**Web** ■ www.cellsignal.com

rev. 01/05/15

**For Research Use Only. Not For Use In Diagnostic Procedures.**

**Entrez-Gene ID** #8350
**UniProt ID** #P68431

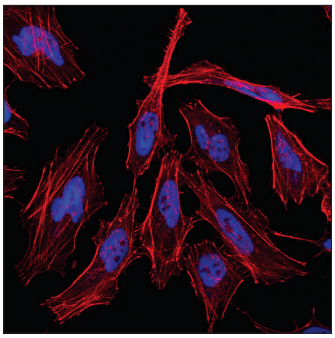
| Applications | Species Cross-Reactivity* | Isotype |
|---|---|---|
| IF-IC, F | H, M, R, Mk, | Rabbit IgG |
| Endogenous | (Hm, C, Dm, X, Z, B) | |

**Description:** This Cell Signaling Technology antibody is conjugated to Alexa Fluor® 647 fluorescent dye and tested in-house for direct flow cytometry and immunofluorescent analysis in human cells. The antibody is expected to exhibit the same species cross-reactivity as the unconjugated Histone H3 (D1H2) XP® Rabbit mAb #4499.

**Background:** Modulation of chromatin structure plays an important role in the regulation of transcription in eukaryotes. The nucleosome, made up of DNA wound around eight core histone proteins (two each of 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, 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, 23, 27, and 56. Acetylation of H3 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 at 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 at Thr3 of H3 in prophase and its dephosphorylation during anaphase (11).

**Specificity/Sensitivity:** Histone H3 (D1H2) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) detects endogenous levels of total histone H3 protein, including the Histone H3 variant CENP-A. This antibody does not cross-react with other core histones.

**Source/Purification:** Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to the carboxy terminus of the human histone H3 protein.



*Confocal immunofluorescent analysis of HeLa cells using Histone H3 (D1H2) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) (blue pseudocolor). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red).*

**Storage:** Supplied in PBS (pH 7.2), less than 0.1% sodium azide and 2 mg/ml BSA. Store at 4°C. *Do not aliquot the antibody. Protect from light. Do not freeze.*

***Species cross-reactivity is determined by western blot using the unconjugated antibody.**

**Recommended Antibody Dilutions:**

| | |
|---|---|
| Immunofluorescence (IF-IC) | 1:800 |
| IF Protocol: | Methanol Permeabilization required |
| Flow Cytometry | 1:50 |

**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.**

**Background References:**

(1) Workman, J.L. and Kingston, R.E. (1998) *Annu Rev Biochem* 67, 545-79.

(2) Hansen, J.C. et al. (1998) *Biochemistry* 37, 17637-41.

(3) Strahl, B.D. and Allis, C.D. (2000) *Nature* 403, 41-5.

(4) Cheung, P. et al. (2000) *Cell* 103, 263-71.

(5) Bernstein, B.E. and Schreiber, S.L. (2002) *Chem Biol* 9, 1167-73.

(6) Jaskelioff, M. and Peterson, C.L. (2003) *Nat Cell Biol* 5, 395-9.

(7) Thorne, A.W. et al. (1990) *Eur J Biochem* 193, 701-13.

(8) Hendzel, M.J. et al. (1997) *Chromosoma* 106, 348-60.

(9) Goto, H. et al. (1999) *J Biol Chem* 274, 25543-9.

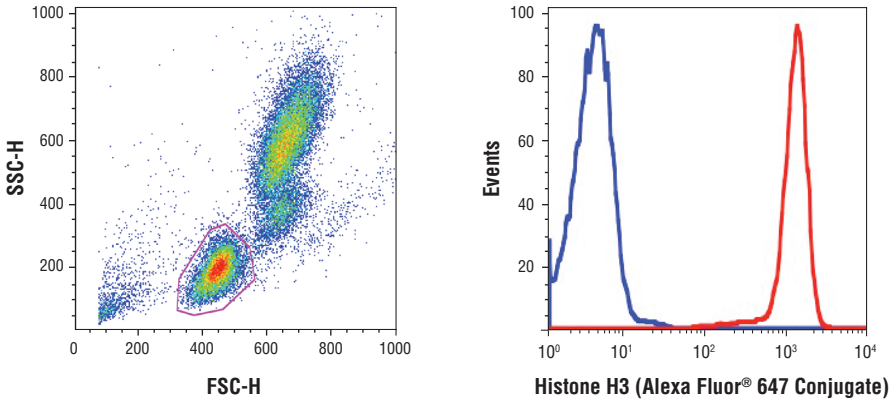
(10) Preuss, U. et al. (2003) *Nucleic Acids Res* 31, 878-85.

(11) Dai, J. et al. (2005) *Genes Dev* 19, 472-88.





**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.




*Human whole blood was fixed, lysed, and permeabilized as per the Cell Signaling Technology Alternate Flow Protocol and stained using Histone H3 (D1H2) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate). The forward/side-scatter lymphocyte gate was applied to a histogram depicting the mean fluorescence intensity of Histone H3 (blue) versus that of concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).*