

Tri-Methyl-Histone H3 (Lys27) (C36B11) Rabbit mAb (Alexa Fluor® 647 Conjugate)

✓ 100 µl
(50 tests)



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

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 Tri-Methyl-Histone H3 (Lys27) (C36B11) Rabbit mAb #9733.

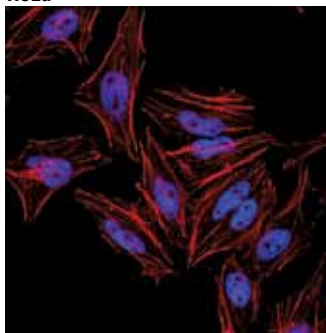
Background: The nucleosome, made up of four core histone proteins (H2A, H2B, H3, and H4), is the primary building block of chromatin. Originally thought to function as a static scaffold for DNA packaging, histones have now been shown to be dynamic proteins, undergoing multiple types of post-translational modifications, including acetylation, phosphorylation, methylation, and ubiquitination (1). Histone methylation is a major determinant for the formation of active and inactive regions of the genome and is crucial for the proper programming of the genome during development (2,3). Arginine methylation of histones H3 (Arg2, 17, 26) and H4 (Arg3) promotes transcriptional activation and is mediated by a family of protein arginine methyltransferases (PRMTs), including the co-activators PRMT1 and CARM1 (PRMT4) (4). In contrast, a more diverse set of histone lysine methyltransferases has been identified, all but one of which contain a conserved catalytic SET domain originally identified in the *Drosophila* Su(var)3-9, Enhancer of zeste, and Trithorax proteins. Lysine methylation occurs primarily on histones H3 (Lys4, 9, 27, 36, 79) and H4 (Lys20) and has been implicated in both transcriptional activation and silencing (4). Methylation of these lysine residues coordinates the recruitment of chromatin modifying enzymes containing methyl-lysine binding modules such as chromodomains (HP1, PRC1), PHD fingers (BPTF, ING2), tudor domains (53BP1), and WD-40 domains (WDR5) (5-8). The discovery of histone demethylases such as PADI4, LSD1, JMJD1, JMJD2, and JHDM1 has shown that methylation is a reversible epigenetic marker (9).

Specificity/Sensitivity: Tri-Methyl-Histone H3 (Lys27) (C36B11) Rabbit mAb (Alexa Fluor® 647 Conjugate) recognizes endogenous levels of histone H3 only when tri-methylated at Lys27. The antibody does not cross-react with non-methylated, mono-methylated or di-methylated Lys27. In addition, the antibody does not cross-react with mono-methylated, di-methylated or tri-methylated histone H3 at Lys4, Lys9, Lys36 or histone H4 at Lys20.

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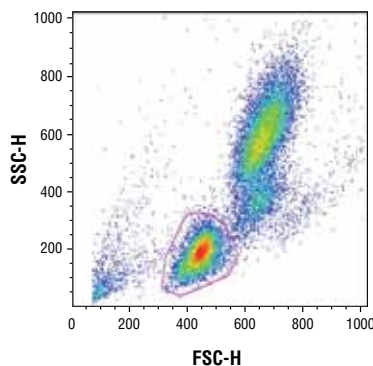
Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E-P—ELISA-Peptide
Species Cross-Reactivity Key: H—human M—mouse R—rat Hm—hamster Mk—monkey Mi—mink C—chicken Dm—D. melanogaster X—Xenopus Z—zebrafish B—bovine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

HeLa



Confocal immunofluorescent analysis of HeLa cells using Tri-Methyl-Histone H3 (Lys27) (C36B11) Rabbit mAb (Alexa Fluor® 647 Conjugate) (blue pseudocolor). Actin filaments were labeled with DyLight™ 554 Phalloidin #13054 (red).

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to the amino terminus of histone H3 in which Lys27 is tri-methylated.



Human whole blood was fixed, lysed, and permeabilized as per the Cell Signaling Technology Flow Alternate Protocol and stained using Tri-Methyl-Histone H3 (Lys27) (C36B11) Rabbit mAb (Alexa Fluor® 647 Conjugate). The forward/side-scatter lymphocyte gate was applied to a histogram depicting the mean fluorescence intensity of Tri-Methyl-Histone H3 (Lys27) (blue) and concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).

Entrez-Gene ID #8350
UniProt ID #P68431

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 antibodies. Protect from light. Do not freeze.

***Species cross-reactivity other than human is determined by western blot using the unconjugated antibody**

Recommended Antibody Dilutions:

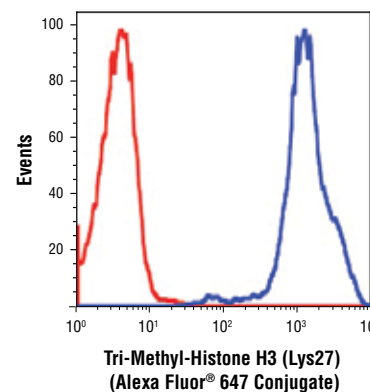
Immunofluorescence (IF-IC)	1:100
Flow Cytometry	1:50

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) Peterson, C.L. and Laniel, M.A. (2004) *Curr. Biol.* 14, R546-R551.
- (2) Kubicek, S. et al. (2006) *Ernst Schering Res. Found Workshop*, 1-27.
- (3) Lin, W. and Dent, S.Y. (2006) *Curr. Opin. Genet. Dev.* 16, 137-142.
- (4) Lee, D.Y. et al. (2005) *Endocr. Rev.* 26, 147-170.
- (5) Daniel, J.A. et al. (2005) *Cell Cycle* 4, 919-926.
- (6) Shi, X. et al. (2006) *Nature* 442, 96-99.
- (7) Wysocka, J. et al. (2006) *Nature* 442, 86-90.
- (8) Wysocka, J. et al. (2005) *Cell* 121, 859-872.
- (9) Trojer, P. and Reinberg, D. (2006) *Cell* 125, 213-217.



U.S. Patent No. 5,675,063
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