Store at 4°C

#12244

Di-Methyl-Histone H3 (Lys27) (D18C8) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate)

☑ 100 µl
(50 tests)

Cell Signaling TECHNOLOGY®

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New 03/13

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

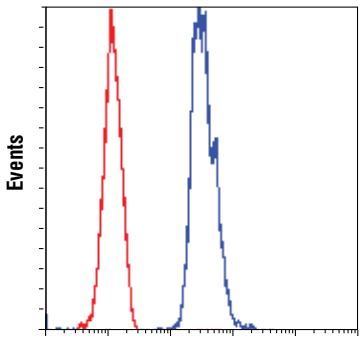
Entrez-Gene ID #8350
Swiss-Prot Acc. #P68431

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

Description: This Cell Signaling Technology antibody is conjugated to Alexa Fluor® 647 fluorescent dye and tested in-house for direct flow cytometric and immunofluorescent analysis in human cells. The antibody is expected to exhibit the same species cross-reactivity as the unconjugated Di-Methyl-Histone H3 (Lys27) (D18C8) XP® Rabbit mAb #9728.

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).

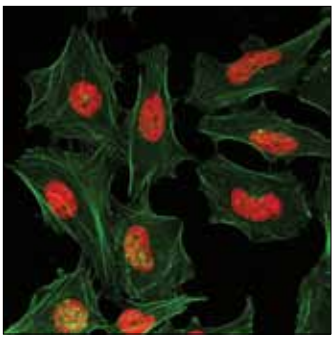
Specificity/Sensitivity: Di-Methyl-Histone H3 (Lys27) (D18C8) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) recognizes endogenous levels of histone H3 when di-methylated at Lys27. The antibody shows some cross-reactivity with mono-methylated Lys27, but does not cross-react with non-methylated or tri-methylated Lys27. In addition, the antibody does not cross-react with mono-methylated, di-methylated or tri-methylated histone H3 Lys4, Lys9, Lys36, or histone H4 Lys20.



Di-Methyl-Histone H3 (Lys27) (Alexa Fluor® 647 Conjugate)

Flow cytometric analysis of Jurkat cells using Di-Methyl-Histone H3 (Lys27) (D18C8) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) (blue) compared to concentration-matched Rabbit (DA1E) mAb IgG XP® Isotype Control (Alexa Fluor® 647 Conjugate) #2985 (red).

HeLa



Confocal immunofluorescent analysis of HeLa cells using Di-Methyl-Histone H3 (Lys27) (D18C8) XP® Rabbit mAb (Alexa Fluor® 647 Conjugate) (red). Actin filaments were labeled with Alexa Fluor® 488 Phalloidin #8878 (green).

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 di-methylated.

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 other than human is determined by western using the unconjugated antibody.**

Recommended Antibody Dilutions:

Immunofluorescence (IF-IC)	1:50
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 complementary 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.

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.