

#13969 Store at -20°C

**Tri-Methyl-Histone H3 (Lys9)  
(D4W1U) Rabbit mAb****Cell Signaling**  
TECHNOLOGY®**Orders:** 877-616-CELL (2355)  
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**For Research Use Only. Not for Use in Diagnostic Procedures.**

Applications:	Reactivity:	Sensitivity:	MW (kDa):	Source/Isotype:	UniProt ID:	Entrez-Gene Id:
WB, IP, IF-IC, FC-FP, ChIP	H M R Mk	Endogenous	17	Rabbit IgG	#P68431	8350

**Product Usage Information**

For optimal ChIP and ChIP-seq results, use 10 µl of antibody and 10 µg of chromatin (approximately 4 x 10<sup>6</sup> cells) per IP. This antibody has been validated using SimpleChIP® Enzymatic Chromatin IP Kits.

**Application****Dilution**

Western Blotting

1:1000

Immunoprecipitation

1:50

Immunofluorescence (Immunocytochemistry)

1:400 - 1:1600

Flow Cytometry (Fixed/Permeabilized)

1:50 - 1:200

Chromatin IP

1:50

**Storage**

Supplied in 10 mM sodium HEPES (pH 7.5), 150 mM NaCl, 100 µg/ml BSA, 50% glycerol and less than 0.02% sodium azide. Store at -20°C. Do not aliquot the antibody.

**Specificity / Sensitivity**

Tri-Methyl-Histone H3 (Lys9) (D4W1U) Rabbit mAb detects endogenous levels of histone H3 when tri-methylated on Lys9. This antibody shows some cross-reactivity with histone H3 that is di-methylated on Lys9, but does not cross-react with non-methylated or mono-methylated histone H3 Lys9. This antibody does not detect tri-methyl histone H3 Lys9 when the adjacent Ser10 residue is phosphorylated during mitosis. In addition, this antibody does not cross-react with methylated histone H3 Lys4, Lys27, Lys36, or Lys79.

**Species predicted to react based on 100% sequence homology:**

Bovine

**Source / Purification**

Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues near the amino terminus of histone H3 in which Lys9 is tri-methylated.

**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).

**Background References**

- Peterson, C.L. and Laniel, M.A. (2004) *Curr Biol* 14, R546-51.
- Kubicek, S. et al. (2006) *Ernst Schering Res Found Workshop*, 1-27.
- Lin, W. and Dent, S.Y. (2006) *Curr Opin Genet Dev* 16, 137-42.
- Lee, D.Y. et al. (2005) *Endocr Rev* 26, 147-70.
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7. Wysocka, J. et al. (2006) *Nature* 442, 86-90.
8. Wysocka, J. et al. (2005) *Cell* 121, 859-72.
9. Trojer, P. and Reinberg, D. (2006) *Cell* 125, 213-7.

<b>Species Reactivity</b>	Species reactivity is determined by testing in at least one approved application (e.g., western blot).
<b>Western Blot Buffer</b>	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.
<b>Applications Key</b>	<b>WB:</b> Western Blotting <b>IP:</b> Immunoprecipitation <b>IF-IC:</b> Immunofluorescence (Immunocytochemistry) <b>FC-FP:</b> Flow Cytometry (Fixed/Permeabilized) <b>ChIP:</b> Chromatin IP
<b>Cross-Reactivity Key</b>	<b>H:</b> human <b>M:</b> mouse <b>R:</b> rat <b>Hm:</b> hamster <b>Mk:</b> monkey <b>Vir:</b> virus <b>Mi:</b> mink <b>C:</b> chicken <b>Dm:</b> D. melanogaster <b>X:</b> Xenopus <b>Z:</b> zebrafish <b>B:</b> bovine <b>Dg:</b> dog <b>Pg:</b> pig <b>Sc:</b> S. cerevisiae <b>Ce:</b> C. elegans <b>Hr:</b> horse <b>GP:</b> Guinea Pig <b>Rab:</b> rabbit <b>All:</b> all species expected
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