

#9758 Store at -20°C

# Di-Methyl-Histone H3 (Lys36) Antibody

✓ 100 µl (10 western blots)



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This product is for *in vitro* research use only and is not intended for use in humans or animals. This product is not intended for use as a therapeutic or in diagnostic procedures.

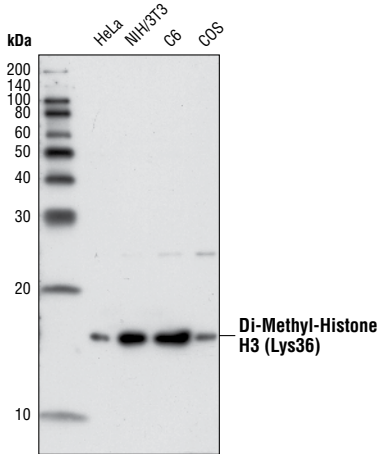
Entrez-Gene ID #8350  
Swiss-Prot Acc. #P68431

| Applications    | Species Cross-Reactivity* | Molecular Wt. | Source   |
|-----------------|---------------------------|---------------|----------|
| W<br>Endogenous | H, M, R, Mk               | 17 kDa        | Rabbit** |

**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 have 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) (5,6,7,8). The recent discovery of histone demethylases such as PADI4, LSD1, JMJD1, JMJD2 and JHDM1 has shown that methylation is a reversible epigenetic mark (9).

**Specificity/Sensitivity:** This antibody detects endogenous levels of histone H3 only when di-methylated on Lys36. The antibody does not cross-react with non-methylated, mono-methylated, or tri-methylated Lys36. In addition, the antibody does not cross-react with di-methylated histone H3 Lys4, Lys9, Lys27, Lys79 or methylated histone H4 Lys20.

**Source/Purification:** Polyclonal antibodies are produced by immunizing animals with a synthetic peptide (KLH-coupled) corresponding to the amino terminus of histone H3 in which lysine 36 is di-methylated. Antibodies are purified by protein A and peptide affinity chromatography.



Western blot analysis of cell lysates from HeLa, NIH/3T3, C6 and COS cells using Di-Methyl Histone H3 (Lys36) Antibody.

### Background References:

- (1) Peterson, C.L. and Lanier, 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.

**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

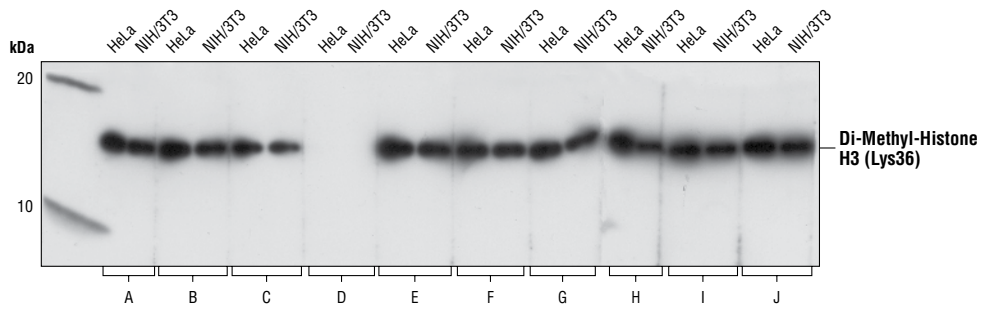
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**IMPORTANT: For Western blots, incubate membrane with diluted antibody in 5% w/v BSA, 1X TBS, 0.1% Tween-20 at 4°C with gentle shaking, overnight.**

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Applications Key: W—Western IP—Immunoprecipitation IHC—Immunohistochemistry ChIP—Chromatin Immunoprecipitation IF—Immunofluorescence F—Flow cytometry E—ELISA 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 All—all species expected Species enclosed in parentheses are predicted to react based on 100% homology.



**Blocking Peptides**

- A None
- B Non-Methyl H3 (Lys36)
- C Mono-Methyl H3 (Lys36)
- D Di-Methyl H3 (Lys36)
- E Tri-Methyl H3 (Lys36)
- F Di-Methyl H3 (Lys4)
- G Di-Methyl H3 (Lys9)
- H Di-Methyl H3 (Lys27)
- I Di-Methyl H3 (Lys79)
- J Di-Methyl H4 (Lys20)

*Antibody specificity was determined by western blotting. HeLa and NIH/3T3 cell lysates were probed with Di-Methyl Histone H3 (Lys36) Antibody (Panel A) or Di-Methyl Histone H3 (Lys36) Antibody pre-adsorbed with 1.5  $\mu$ M of various competitor peptides (Panels B-J). As shown, only the di-methyl histone H3 (Lys36) peptide competed away binding of the antibody.*