#12522 Store at -20°C

Mono-Methyl-Histone H3 (Lys79) (D5X1S) Rabbit mAb

☑ 100 µl
(10 western blots)

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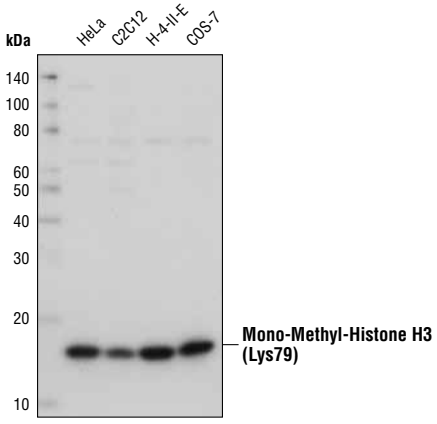
Entrez-Gene ID #8350
Swiss-Prot Acc. #P68431

Applications	Species Cross-Reactivity*	Molecular Wt.	Isotype
W, ChIP	H, M, R, Mk	17 kDa	Rabbit IgG**
Endogenous			

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: Mono-Methyl-Histone H3 (Lys79) (D5X1S) Rabbit mAb recognizes endogenous levels of histone H3 protein only when mono-methylated at Lys79. This antibody does not cross-react with non-methylated, di-methylated, or tri-methylated histone H3 Lys79. In addition, the antibody does not cross-react with histone H3 mono-methylated at Lys4, Lys9, Lys27, or Lys36.

Source/Purification: Monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to residues surrounding mono-methyl Lys79 of human histone H3 protein.



Western blot analysis of extracts from various cell lines using Mono-Methyl-Histone H3 (Lys79) (D5X1S) Rabbit mAb.

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.*

***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
Chromatin IP	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.

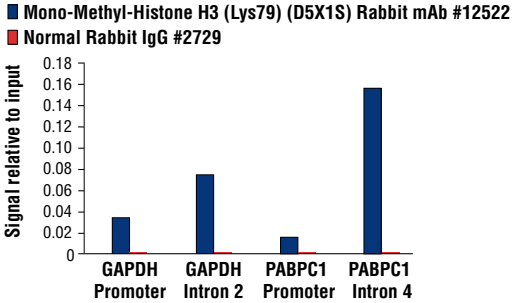
(5) Daniel, J.A. et al. (2005) *Cell Cycle* 4, 919-926.

(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.

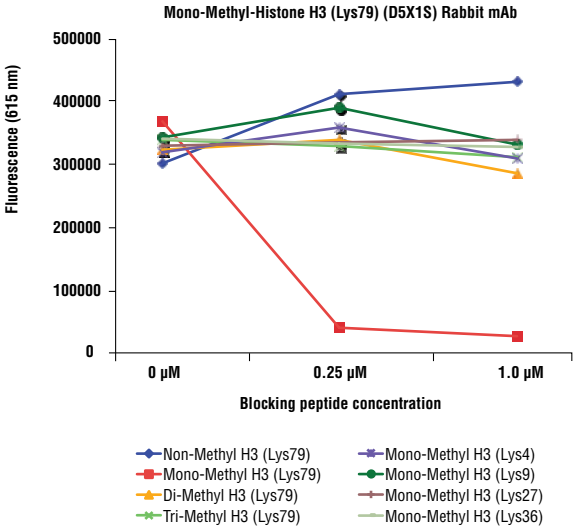
(9) Trojer, P. and Reinberg, D. (2006) *Cell* 125, 213-217.



Chromatin immunoprecipitations were performed with cross-linked chromatin from 4 x 10^6 HeLa cells and either 10 µl of Mono-Methyl-Histone H3 (Lys79) (D5X1S) Rabbit mAb or 2 µl of Normal Rabbit IgG #2729 using SimpleChIP® Enzymatic Chromatin IP Kit (Magnetic Beads) #9003. The enriched DNA was quantified by real-time PCR using SimpleChIP® Human GAPDH Promoter Primers #4471, SimpleChIP® Human GAPDH Intron 2 Primers #4478, human PABPC1 promoter primers, and human PABPC1 intron 4 primers. The amount of immunoprecipitated DNA in each sample is represented as signal relative to the total amount of input chromatin, which is equivalent to one.

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.

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.



Mono-Methyl Histone H3 (Lys79) (D5X1S) Rabbit mAb specificity was determined by peptide ELISA. The graph depicts the binding of the antibody to pre-coated mono-methyl histone H3 (Lys79) peptide in the presence of increasing concentrations of various competitor peptides. As shown, only the mono-methyl histone H3 (Lys79) peptide competed away binding of the antibody.