Store at -20°C

Methyl-Histone H3 (Lys9) Antibody Sampler Kit

#76252

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New 04/18

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

Products Included	Product #	Quantity	Mol.Wt.	Isotype/Source
Tri-Methyl-Histone H3 (Lys9) (D4W1U) Rabbit mAb	13969	20 µl	17 kDa	Rabbit IgG
Di-Methyl-Histone H3 (Lys9) (D85B4) XP® Rabbit mAb	4658	20 µl	17 kDa	Rabbit IgG
Mono-Methyl-Histone H3 (Lys9) (D1P5R) Rabbit mAb	14186	20 µl	17 kDa	Rabbit IgG
Histone H3 (D1H2) XP® Rabbit mAb	4499	20 µl	17 kDa	Rabbit IgG
Anti-Rabbit IgG, HRP-linked Antibody	7074	100 µl		Goat

See www.cellsignal.com for individual component applications, species cross-reactivity, dilutions and additional application protocols.

Description: The Methyl-Histone H3 (Lys9) Antibody Sampler Kit provides an economical means of detecting levels of mono-, di-, and tri-methyl histone H3 Lys9 using methyl-specific and control histone H3 antibodies. The kit contains enough primary antibodies to perform at least two western blot experiments.

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).

Methylation of histone H3 Lys9 is generally associated with transcriptional repression of constitutive heterochromatin. Tri- and di-methyl-histone H3 Lys9 levels are high in regions of transcriptionally repressed facultative heterochromatin, including transposable elements and centromeric regions. Mono-methyl-histone H3 Lys9 levels are more widely dispersed and found in the bodies of active and inactive genes.

Specificity/Sensitivity: Each antibody in the Methyl-Histone H3 (Lys9) Antibody Sampler Kit detects endogenous levels of its target protein. 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. Di-Methyl-Histone H3 (Lys9) (D85B4) XP® Rabbit mAb detects endogenous levels of histone H3 only when di-methylated on Lys9. The antibody does not cross-react with non-methylated, mono-methylated or tri-methylated Histone H3 Lys9. Mono-Methyl-Histone H3 (Lys9) (D1P5R) Rabbit mAb recognizes endogenous levels of histone H3 protein only when mono-methylated at Lys9. This antibody does not cross-react with non-methylated, di-methylated, or tri-methylated Lys9. Histone H3 (D1H2) XP® Rabbit mAb detects endogenous levels of total Histone H3 protein, including isoforms H3.1, H3.2, H3.3, and the variant histone CENP-A. This antibody does not cross-react with other core histones.

Source/Purification: Monoclonal methyl-histone H3 Lys9 antibodies are produced by immunizing rabbits with synthetic peptides corresponding to the amino terminus of histone H3 in which Lys9 is mono-, di-, or tri-methylated. The control histone H3 monoclonal antibody is produced by immunizing animals with a synthetic peptide corresponding to the carboxy terminus of the human histone H3 protein.

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 antibodies.*

Background References:

(1) Peterson, C.L. and Laniel, M.A. (2004) *Curr Biol* 14, R546-51.

(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, 137-42.

(4) Lee, D.Y. et al. (2005) *Endocr Rev* 26, 147-70.

(5) Daniel, J.A. et al. (2005) *Cell Cycle* 4, 919-26.

(6) Shi, X. et al. (2006) *Nature* 442, 96-9.

(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.

U.S. Patent No. 7,429,487, foreign equivalents, and child patents deriving therefrom.

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Applications: **W**—Western **IP**—Immunoprecipitation **IHC**—Immunohistochemistry **ChIP**—Chromatin Immunoprecipitation **IF**—Immunofluorescence **F**—Flow cytometry **E-P**—ELISA-Peptide **Species Cross-Reactivity:** **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.