

Histone H3K9me2 antibody (mAb)

Catalog No: 39683

RRID: AB_2793304

Clone: MABI 0307

Isotype: IgG1

Application(s): ChIP, DB, ICC, IF, WB

Reactivity: Human, Wide Range Predicted

Quantity: 100 µg

Purification: Protein G Chromatography

Host: Mouse

Concentration: 0.8 µg/µl

Molecular Weight: 17 kDa

Background: Histone H3 is one of the core components of the nucleosome. The nucleosome is the smallest subunit of chromatin and consists of 147 base pairs of DNA wrapped around an octamer of core histone proteins (two each of Histone H2A, Histone H2B, Histone H3 and Histone H4). Histone H1 is a linker histone, present at the interface between the nucleosome core and DNA entry/exit points. Histone H1 is responsible for establishing higher-order chromatin structure. Chromatin is subject to a variety of chemical modifications, including post-translational modifications of the histone proteins and the methylation of cytosine residues in the DNA. Reported histone modifications include acetylation, methylation, phosphorylation, ubiquitylation, glycosylation, ADP-ribosylation, carbonylation and SUMOylation; these modifications play a major role in regulating gene expression.

The methylation of histones can occur on two different residues: arginine or lysine. Histone methylation can be associated with transcriptional activation or repression, depending on the methylated residue. Lysine 9 of histone H3 can be mono-, di- or trimethylated by different histone methyltransferases (HMTs) such as SuvH39H1 or G9a. This methylated lysine can be demethylated by histone demethylases as JMJD1A, LSD1 or JMJD2C. Methylation of this residue is mainly associated with transcriptional repression.

Immunogen: This Histone H3 dimethyl Lys9 antibody was raised against a synthetic peptide containing dimethyl Lys9 of human histone H3.

Buffer: PBS pH 7.5 containing 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.

Application Notes:

Applications Validated by Active Motif:

ChIP: 10 µg per ChIP

WB: 0.5 - 2 µg/ml dilution

ICC/IF: 1 - 2 µl/ml dilution

Storage and Guarantee: Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product is guaranteed for 12 months from date of receipt.

This product is for research use only and is not for use in diagnostic procedures.

This antibody is manufactured by MAB Institute, Inc.

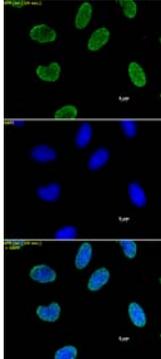


1 2 3

Histone H3 dimethyl Lys9 antibody tested by ChIP.

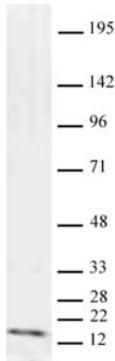
Chromatin IP performed using the ChIP-IT Express Kit (Catalog No. 53008) and 100 µl of Ready-to-ChIP HeLa Chromatin (Catalog No. 53015) per ChIP. Subsequent to the ChIP reaction, DNA was purified from the immunoprecipitated chromatin and a region 1 kb upstream of the human HOXD13 promoter was amplified by PCR.

- Lane 1: ChIP using 10 µg of Histone H3 dimethyl Lys9 antibody.
- Lane 2: PCR input control.
- Lane 3: ChIP using negative control mouse IgG.



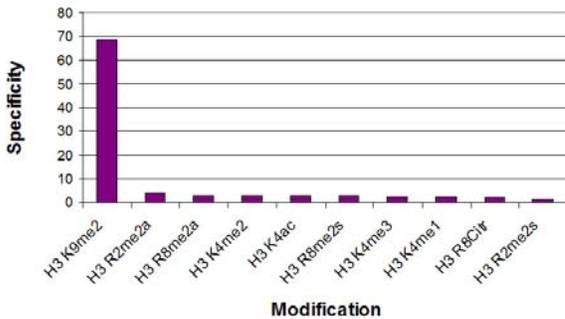
Histone H3 dimethyl Lys9 antibody tested by immunofluorescence.

Staining of HeLa cells with Histone H3 dimethyl Lys9 antibody (1 µg/ml, top panel) and DAPI (middle panel), and a merge of both images (bottom panel).



Histone H3 dimethyl Lys9 antibody tested by Western blot.

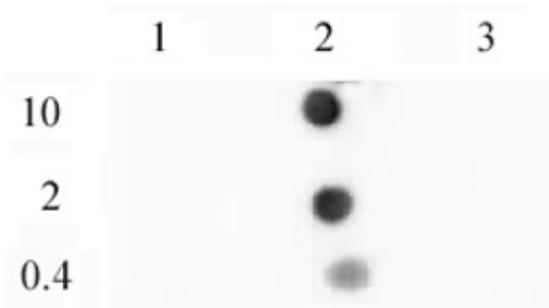
HeLa nuclear extract (20 µg) probed with Histone H3 dimethyl Lys9 antibody (2 µg/ml dilution).



Histone H3 dimethyl Lys9 antibody specificity tested by peptide array analysis.

Peptide array analysis was used to confirm the specificity of this antibody for its intended modification. Histone H3 dimethyl Lys9 antibody was applied at a dilution of 0.1 µg / ml to Active Motif's MODified™ Histone Peptide Array (Catalog No. 13001). The arrays were scanned with ArrayAnalysis Software 7 and the results plotted. Specificity data is shown for the most reactive peptides and those related to the immunogen. Recognition of the H3 dimethyl Lys9 peptide is inhibited by dimethylation at Arg8 and blocked by Ser10 and Thr11 phosphorylation and by Lys14 acetylation.

Array Data File



Histone H3 dimethyl Lys9 mAb tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H3 dimethyl Lys9 mAb for dimethyl-Lys9 histone H3. Recombinant methylated proteins corresponding to the immunogen and related proteins were spotted onto PVDF and probed with the antibody at 0.3 µg/ml. The amount of protein (picomoles) spotted is indicated next to each row.

- Lane 1: monomethyl lysine 9 protein.
- Lane 2: dimethyl lysine 9 protein.
- Lane 3: trimethyl lysine 9 protein.

No detection of proteins (unmodified, mono-, di-, or tri-methylated) corresponding to lysine 27 of histone H3 was observed with Histone H3 dimethyl Lys9 antibody.