

# Histone H3K27me2 antibody (mAb)

Catalog No: 61435 RRID: AB 2793635

Clone: MABI 0324

Application(s): ChIP, ChIP-Seq, CUT&Tag, WB

Reactivity: Human, Wide Range Predicted

**Quantity**: 100 μg **Purification**: None

Host: Mouse Isotype: IgG2a

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; it 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; they play a major role in regulating gene expression.

Histone H3K27 can be mono-, di- or trimethylated by different histone methyltransferases, such as EZH2 or NSD3. While histone methylation can be associated with transcriptional activation or repression, methylation of Lysine 27 of histone H3 is mainly associated with transcriptional repression.

Immunogen: This antibody was raised against a synthetic peptide containing dimethyl Lysine 27 of human histone H3.

Buffer: PBS pH 7.5 containing 30% glycerol. 0.3M NaCl, and 0.035% sodium azide. Sodium azide is highly toxic.

## **Application Notes:**

Applications Validated by Active Motif:

ChIP: 2 µg per ChIP

WB\*: 0.5 - 2 µg/ml dilution

CUT&Tag: 1-2 µg per 50 µl reaction

\*Note: many chromatin-bound proteins are not soluble in a low salt nuclear extract and fractionate to the pellet. Therefore, we recommend a High Salt / Sonication Protocol when preparing nuclear extracts for Western Blot.

**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.

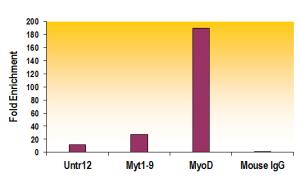
This antibody is manufactured by MAB Institute, Inc.





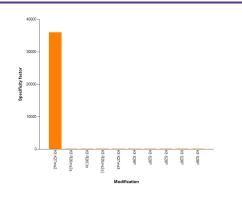
### Histone H3K27me2 antibody (mAb) tested by ChIP.

Chromatin IP performed using the ChIP-IT® Express Kit (Catalog No. 53008) and HeLa Chromatin (1.5 x 10^6 cell equivalents per ChIP) using 2 µg of Histone H3K27me2 antibody or the equivalent amount of IgG as a negative control. Real time, quantitative PCR (RT-qPCR) was performed on DNA purified from the ChIP reaction using primer pairs specific for the Myt1-9 gene, the MyoD gene or the negative control primer pair Untr12. Data are presented as Fold Enrichment of the ChIP antibody signal versus the negative control IgG using the ddCT method.



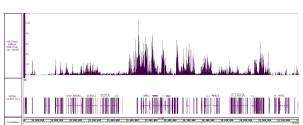
### Histone H3K27me2 antibody (mAb) tested by Western blot.

Nuclear extract of HeLa cells (30  $\mu g$ ) probed with Histone H3K27me2 antibody (mAb) at a dilution of 2  $\mu g$ /ml.



#### Histone H3K27me2 antibody (mAb) specificity tested by peptide array analysis.

Peptide array analysis was used to confirm the specificity of this antibody for its intended modification. Histone H3K27me2 antibody was applied at a dilution of 0.2 μg/ml to Active Motif's MODified<sup>™</sup> 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.



## Histone H3K27me2 antibody (mAb) tested by CUT&Tag

CUT&Tag was performed using 100,000 K562 cells and sequenced using 38 base-pair, paired-end reads on the Illumina NovaSeq. Data was collected from 6 million reads, and H3K27me2 data is shown for Chromosome 22.