

Histone H3K27me2 antibody (pAb)

Catalog Nos: 39245, 39246

RRID: AB_2793202

Application(s): ChIP, ChIP-Seq, DB, ICC, IF, WB **Reactivity:** Human, Wide Range Predicted

Volumes: 100 μl, 10 μl Purification: None

Host: Rabbit **Isotype:** Serum

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 Histone H3 dimethyl Lys27 antibody was raised against a peptide including dimethyl-lysine 27 of histone H3.

Buffer: Rabbit serum containing 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic. For your convenience, an IgG version (Catalog No. 39919) of this antibody that was purified by Protein A Chromatography is also available.

Application Notes:

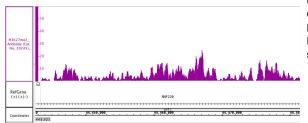
Applications Validated by Active Motif:

ChIP-Seq: 4-6 µl per ChIP ChIP: 5 - 10 µl per ChIP ICC/IF: 1:200 - 1:500 dilution WB: 1:500 - 1:1,000 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.





Histone H3K27me2 antibody (pAb) tested by ChIP-Seq.

Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT $^{\otimes}$ High Sensitivity Kit (Cat. No. 53040) with 30 μg of chromatin from lung tumor chromatin and 6 μl Histone H3K27me2 antibody. ChIP DNA was sequenced on the Illumina HiSeq and 9.1 million sequence tags were mapped to identify Histone H3K27me2 binding sites.



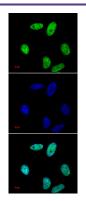
Histone H3 dimethyl Lys27 antibody tested by ChIP.

Chromatin IP performed using the ChIP-IT® Express Kit (Catalog No. 53008) and 50 μ I 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 approximately 1000 base pairs 5' of the transcriptional start site of the HoxD13 gene was amplified by PCR.

Lane 1: ChIP using negative control rabbit IgG.

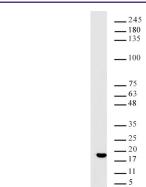
Lane 2: ChIP using 10 µl of Histone H3 dimethyl Lys27 antibody.

Lane 3: PCR input control.



Histone H3 dimethyl Lys27 antibody tested by immunofluorescence.

Staining of HeLa cells with Histone H3 dimethyl Lys27 antibody (1:500 dilution, top panel) and DAPI (middle panel), and a merge of both images (bottom panel).



Histone H3 dimethyl Lys27 antibody tested by Western blot.

Detection of Histone H3 dimethyl Lys27 by Western blot. The analysis was performed using Histone H3 dimethyl Lys27 antibody at a 1:1,000 dilution. HeLa acid extract (20 µg per lane).



Histone H3 dimethyl Lys27 antibody tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H3 dimethyl Lys27 antibody for dimethyl Lys27 histone H3. Methylated peptides corresponding to the immunogen and related sequences derived from histone H3 were spotted onto PVDF and probed with the antibody at a 1:5,000 dilution. The amount of peptide (picomoles) spotted is indicated next to each row. Lane 1: Unmodified lysine 4 peptide. Lane 2: Monomethyl lysine 4 peptide. Lane 3: Dimethyl lysine 4 peptide. Lane 4: Trimethyl lysine 4 peptide. Lane 5: Unmodified lysine 9 peptide. Lane 6: Monomethyl lysine 9 peptide. Lane 7: Dimethyl lysine 9 peptide. Lane 8: Trimethyl lysine 9 peptide. Lane 9: Unmodified lysine 27 peptide. Lane 10: Monomethyl lysine 27 peptide. Lane 11: Dimethyl lysine 27 peptide. Lane 12: Trimethyl lysine 27 peptide.