

Histone H3K27me3 antibody (pAb)

Catalog Nos: 39156, 39158

RRID: AB_2636821

Application(s): ChIP, CUT&Tag, DB, ICC, IF, IP, WB

Reactivity: Human, Mouse, 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 trimethyl Lys27 antibody was raised against a peptide including trimethyl-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. 39155) of this antibody that was purified by Protein A Chromatography is also available.

Application Notes:

Applications Validated by Active Motif:

ChIP: 10 µl per ChIP

ICC/IF: 1:1,000 - 1:2,500 dilution

WB*: 1:1,000 - 1:5,000 dilution

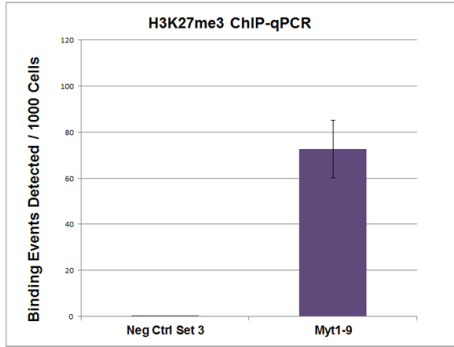
CUT&Tag: 1 µl per 50 µl reaction

*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.

For H3K27me3, we also offer AbFlex® H3K27me3 Recombinant Antibody (rAb). For details, see Catalog No. 91167.

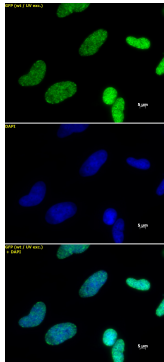
Storage and Guarantee: Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product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

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



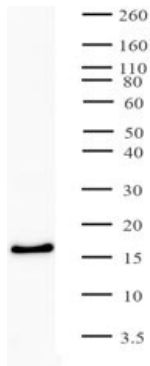
Histone H3K27me3 antibody (pAb) tested by ChIP.

Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT[®] High Sensitivity Kit (Cat. No. 53040) with 15 µg of chromatin from HeLa cells and 5 µl H3K27me3 antibody. ChIP DNA was used in qPCR with the control primer pairs or gene-specific primer pairs as indicated. Data are presented as Binding Events Detected per 1000 Cells using Active Motif's Epigenetic Services normalization scheme which accounts for primer efficiency and the amount of chromatin used in the ChIP reaction.



Histone H3K27me3 antibody (pAb) tested by immunofluorescence.

Staining of HeLa cells with Histone H3 trimethyl Lys27 antibody (1:1,000, top panel) and DAPI (middle panel), and a merge of both images (bottom panel).



Histone H3K27me3 antibody (pAb) tested by Western blot.

HeLa nuclear extract (20 µg) was probed with Histone H3 trimethyl Lys27 antibody (1:1,000 dilution).

Histone H3K27me3 antibody (pAb) tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H3 trimethyl Lys27 antibody for trimethyl-lysine 27 of histone H3. Peptides corresponding to regions around major sites of histone H3 methylation (lysine 4, lysine 9, lysine 27) were spotted onto PVDF and probed with the antibody at a dilution of 1:5,000. The amount of peptide (in picomoles) spotted is indicated next to each row.



Lane 1: Unmodified Lys4 peptide. Lane 2: Monomethyl Lys4 peptide. Lane 3: Dimethyl Lys4 peptide. Lane 4: Trimethyl Lys4 peptide. Lane 5: Monomethyl Lys9 peptide. Lane 6: Unmodified Lys9 peptide. Lane 7: Dimethyl Lys9 peptide. Lane 8: Trimethyl Lys9 peptide. Lane 9: Unmodified Lys27 peptide. Lane 10: Monomethyl Lys27 peptide. Lane 11: Dimethyl Lys27 peptide. Lane 12: Trimethyl Lys27 peptide.

Histone H3K27me3 antibody (pAb) (Cat. 39156) tested by CUT&Tag

CUT&Tag was performed using 100,000 cells and sequenced using 38 base-pair, paired-end reads on the Illumina NovaSeq. Data was collected from 36 million reads, and H3K27me3 data is shown for Chromosome 2.

