Histone H3K27ac antibody (pAb)

Catalog Nos: 39133, 39034, 39134

Isotype: IgG
Application(s): ChIP, ChIP-Seq, CUT&Tag, DB, ICC, IF, IHC, WB
Reactivity: Budding Yeast, Human, Wide Range Predicted

Quantities: 100 µg, 50 µg, 10 µg
Purification: Protein A Chromatography
Host: Rabbit
Concentration: 1 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; 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. Lysine N-ε-acetylation is a dynamic, reversible and tightly regulated protein and histone modification that plays a major role in chromatin remodeling and in the regulation of gene expression in various cellular functions. Acetylation of histone H3 occurs at several different lysine positions in the histone tail, and is performed by Histone Acetyltransferases (HATs) such as CBP/p300. Acetylation of histone H3 at Lys27 is associated with transcriptional activation. Histone H3K27 can also 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 acetyl Lys27 antibody was raised against a peptide including acetyl-lysine 27 of histone H3.

Buffer: Purified IgG in PBS with 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic. For your convenience, an unpurified serum version (Catalog No. 39135) of this antibody is also available.

Application Notes:
Applications Validated by Active Motif:
ChIP: 10 µg per ChIP
ChIP-Seq: 5 µg each
ICC/IF: 1 - 5 µg/ml dilution
WB*: 0.1 - 1 µg/ml dilution
CUT&Tag: 1 µg per 50 µl reaction*

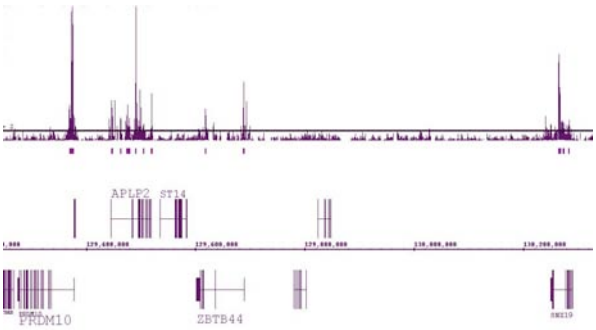
*This antibody has been validated for CUT&Tag using Active Motif's CUT&Tag-IT™ Assay Kit, Catalog No. 53160.

*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.

modENCODE validation: this antibody was validated for ChIP-Seq in this study (see references).

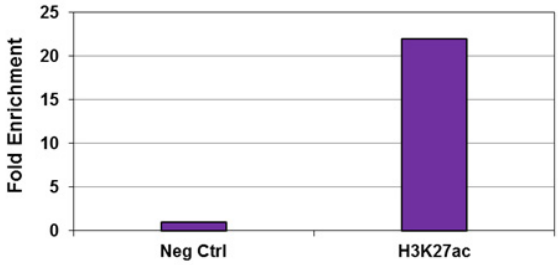
NGS-QC® certification: this antibody has been processed by the NGS-QC® generator.

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.



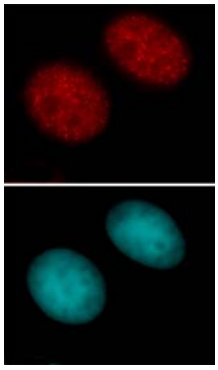
Histone H3K27ac antibody (pAb) tested by ChIP-Seq

Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) with 30 µg of HAP1 myeloid leukemia cell chromatin and 4 µg of Histone H3K27ac antibody. ChIP DNA was sequenced on the Illumina NextSeq and 17 million sequence tags were mapped to identify Histone H3K27ac binding sites on chromosome 2.



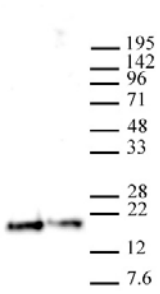
Histone H3 acetyl Lys27 antibody tested by ChIP qPCR analysis.

ChIP was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) and HeLa Chromatin (1.5 x 10^6 cell equivalents per ChIP) using 10 µg of Histone H3K27ac pAb or rabbit IgG as a negative control. Real time, quantitative PCR (RT-qPCR) was performed on DNA purified from each of the ChIP reactions using a Human Positive Control Primer Set GAPDH-2 (Cat. No. 71006). Data are presented as Fold Enrichment of the ChIP antibody signal versus the negative control IgG using the ddCT method.



Histone H3 acetyl Lys27 antibody tested by immunofluorescence.

Staining of HEK293 cells with Histone H3 acetyl Lys27 antibody (1 µg/ml dilution, top panel) and DAPI (bottom panel).

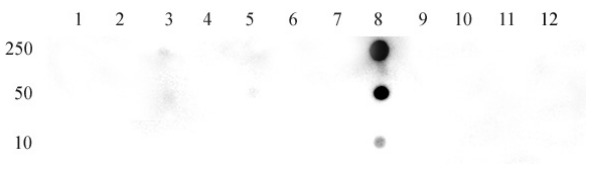


Histone H3K27ac antibody tested by Western blot.

20 µg of HeLa cell nuclear extract was probed with Histone H3K27ac polyclonal antibody at 0.2 µg/ml

Lane 1: Cells treated with sodium butyrate..

Lane 2: No treatment.



Histone H3K27ac antibody tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H3K27ac antibody. Acetylated peptides were spotted onto PVDF and probed with antibody at 1 µg/ml. The amount of peptide (picomoles) spotted is indicated on the left.

Column 1: H3K37ac. Column 2: H3K36ac. Column 3: H3K9ac. Column 4: H3K14ac. Column 5: H3K18ac. Column 6: H3K23ac. Column 7: unmod H3K27. Column 8: H3K27ac. Column 9: H4K5ac. Column 10: H4K8ac. Column 11: H4K12ac. Column 12: H4K16ac.