

## Histone H4K5ac antibody (pAb)

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**Catalog Nos:** 39169, 39170

**RRID:** AB\_2793171

**Isotype:** Serum

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

**Reactivity:** Human, Wide Range Predicted

**Volumes:** 200 µl, 10 µl

**Purification:** None

**Host:** Rabbit

**Molecular Weight:** 8 kDa

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**Background:** Histone H4 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. Histone H4 molecules acetylated at Lys5 or Lys8 are distributed in overlapping, but non-identical, islands throughout the euchromatic chromosome arms.

**Immunogen:** This Histone H4 acetyl Lys5 antibody was raised against a peptide including acetyl-lysine 5 of histone H4.

**Buffer:** Rabbit serum containing 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.

**Application Notes:**

Applications Validated by Active Motif:

ChIP: 3 - 10 µl per ChIP

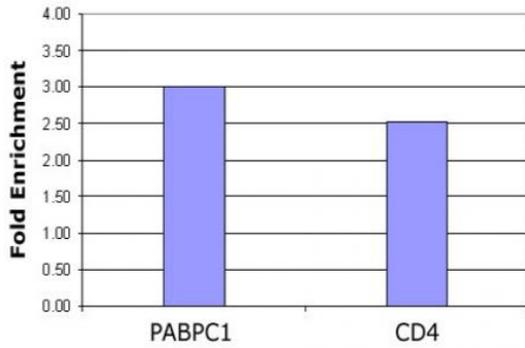
ICC/IF: 1:500 - 1:1,000 dilution

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

\*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.

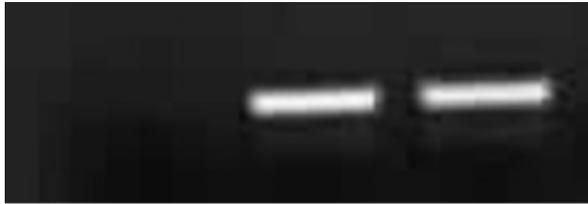
**Storage and Guarantee:** Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This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



**Histone H4 acetyl Lys5 antibody tested by ChIP analysis.**

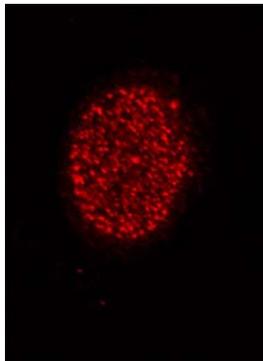
Chromatin IP performed using the ChIP-IT<sup>®</sup> Express Kit (Catalog No. 53008) and HeLa Chromatin (1.5 x 10<sup>6</sup> cell equivalents per ChIP) using 10 µl of Histone H4 acetyl Lys5 antibody or the equivalent amount of rabbit IgG as a negative control. Real time, quantitative PCR (RT-qPCR) was performed on DNA purified from each of the ChIP reactions using a primer pair specific for the indicated gene. Data are presented as Fold Enrichment of the ChIP antibody signal versus the negative control IgG using the ddCT method.



**Histone H4 acetyl Lys5 antibody tested by ChIP analysis.**

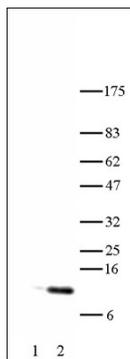
Chromatin IP performed on HeLa cell chromatin using 39169. PCR was performed using primers specific for the promoter region of the human GAPDH gene. Lane 1: negative IgG control. Lane 2: ChIP using 10 ul of 39169. Lane 3: Input DNA control.

1 2 3



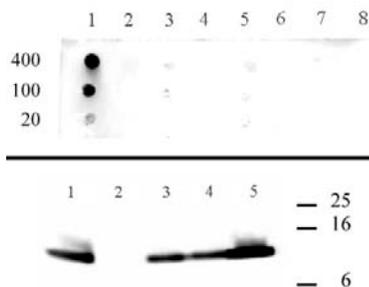
**Histone H4 acetyl Lys5 antibody tested by immunofluorescence.**

Staining of HeLa cells with Histone H4 acetyl Lys5 antibody (1:500 dilution, top panel) and DAPI (bottom panel).



**Histone H4 acetyl Lys5 antibody tested by Western blot.**

HeLa acid extract probed with Histone H4 acetyl Lys5 polyclonal antibody (1:1,000 dilution).  
 Lane 1: No treatment.  
 Lane 2: Cells treated with sodium butyrate.



**Histone H4 acetyl Lys5 antibody specificity data.**

Top Panel – Dot blot analysis was used to confirm the specificity of Histone H4 acetyl Lys5 antibody for H4 acetyl lysine 5. Peptides corresponding to regions around major sites of histone H4 acetylation were spotted onto PVDF and probed at a dilution of 1:1,000. The amount of peptide (in picomoles) spotted is indicated next to each row. Lane 1: acetyl-Lys5 peptide. Lane 2: unmodified Lys5 peptide. Lane 3: acetyl-Lys8 peptide. Lane 4: unmodified Lys8 peptide. Lane 5: acetyl-Lys12 peptide. Lane 6: unmodified Lys12 peptide. Lane 7: acetyl-Lys16 peptide. Lane 8: unmodified Lys16 peptide.

Bottom Panel – Peptide inhibition analysis. A Western blot was performed on butyrate-treated HeLa cell acid extract (5 µl per lane) at a dilution of 1:2,000. Different peptides were incubated with the antibody during the blotting procedure to determine if the recognition was blocked by a specific modification. Lane 1: no peptide. Lane 2: acetyl-Lys5 peptide. Lane 3: acetyl-Lys8 peptide. Lane 4: acetyl-Lys12 peptide. Lane 5: acetyl-Lys16 peptide.