# Histone H2A.Z antibody (pAb)



## Catalog Nos: 39113, 39013, 39114 RRID: AB\_2615081

Application(s): ChIP, ChIP-Seq, DB, IF, WB Reactivity: Human, Wide Range Predicted Volumes: 100 µl, 50 µl, 10 µl Purification: None Host: Rabbit Isotype: Serum Molecular Weight: 14 kDa

**Background:** 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 H2A.Z (H2AZ, H2AFZ) is a histone H2A variant, a protein similar to canonical H2A but with different molecular identity and unique functions. H2A.Z is highly conserved during evolution. It plays an important role in basic cellular mechanisms such as gene activation, chromosome segregation, heterochromatic silencing and progression through the cell cycle. H2A.Z is acetylated at multiple lysine residues in its amino terminus, which may enable H2A.Z to function as an insulator of chromatin domains.

**Immunogen:** This Histone H2A.Z antibody was raised against a peptide derived from the C-terminus of human histone H2A. Z.

**Buffer:** Rabbit serum containing 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic. For your convenience, an IgG version (Catalog No. 39943) of this antibody that was purified by Protein A Chromatography is also available.

## **Application Notes:**

Applications Validated by Active Motif: ChIP: 2 - 10 µl per ChIP ChIP-Seq: 5 µl each WB\*: 1:1,000 - 1:5,000 dilution CUT&Tag: 1 µl per 50 µl reaction

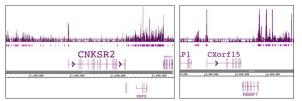
This histone variant can be perturbed by excessive washing steps prior to fixation. Cells should be washed into serum-free culture medium (NOT PBS!) prior to fixation.

\*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.

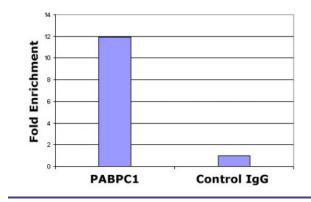
Application Key: ChIP = Chromatin Immunoprecipitation; FACS = Flow Cytometry; IF = Immunofluorescence; IHC = Immunohistochemistry; IP = Immunoprecipitation; WB = Western Blot





#### Histone H2A.Z antibody tested by ChIP-Seq.

ChIP was performed using the ChIP-IT<sup>®</sup> High Sensitivity Kit (Cat. No. 53040) with chromatin from the human H9 embryonic stem cell line (4.5 million cells) and 5 ul of antibody. ChIP DNA was sequenced on the Illumina HiSeq and 30 million sequence tags were mapped to identify H2A.Z occupancy. H2A.Z is found throughout the genome, is often enriched at promoters and depleted from transcribed genes as shown in the images.



25 20 17

\_ 5

### Histone H2A.Z 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  $\mu$ I of Histone H2A.Z pAb or 10  $\mu$ I 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 H2A.Z antibody tested by Western blot.

Detection of H2A.Z by Western blot. The analysis was performed using20 µg of HeLa cell nuclear extract, detected with Histone H2A.Z pAb at a 1:1,000 dilution.

#### Histone H2A.Z antibody tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H2A.Z antibody for histone H2A. Z. Recombinant proteins corresponding to the immunogen and related proteins were spotted onto PVDF and probed with the antibody at 1:4,000. The amount of peptide (picomoles) spotted is indicated next to each row.

Lane 1: recombinant Histone H2A. Lane 2: recombinant Histone H2A.Z. Lane 3: recombinant Histone H4.