Histone H3K79me2 antibody (pAb)

Catalog Nos: 39143, 39144
RRID: AB_2561018
Isotype: Serum
Application(s): ChIP, ChIP-Seq, DB, WB
Reactivity: Human, Wide Range Predicted

Volumes: 100 µl, 10 µl
Purification: None
Host: Rabbit
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). 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; these modifications play a major role in regulating gene expression.

Lysine 79 of histone H3 can be mono-, di- or trimethylated by Dot1 methylase; methylation at this residue acts as a marker of inactive chromatin regions that is critical for transcriptional silencing, and it is thought that silencing proteins such as Sir3 function by blocking Dot1 methylation.

Immunogen: This Histone H3 dimethyl Lys79 antibody was raised against a peptide including dimethyl-lysine 79 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. 39923) of this antibody that was purified by Protein A Chromatography is also available.

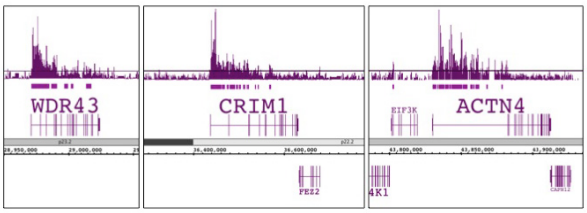
Application Notes:
Applications Validated by Active Motif:
ChIP: 10 µl per ChIP
ChIP-Seq: 5 µl each
WB*: 1:500 - 1:5,000 dilution

The modENCODE and NIH Roadmap Epigenomics Mapping Consortiums have implemented rigorous standardization criteria for all assays and reagents to be used. As part of this initiative, antibody specificity testing and the ability of the antibodies to work in ChIP-Seq were assessed in a large-scale study. This Histone H3 dimethyl Lys79 antibody was validated for ChIP-Seq in the study (see reference).

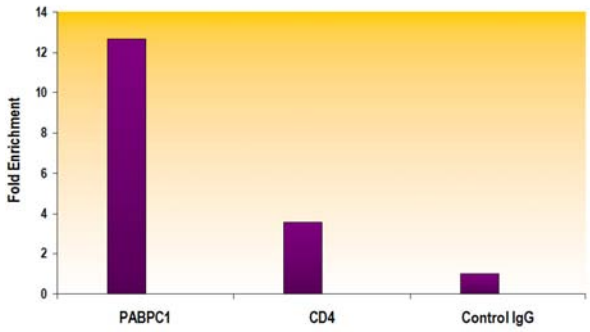
*Note: many chromatin-bound proteins are not soluble in a low salt nuclear extract and fractionate to the pellet. Therefore, we recommend a High Salt / Sonication Protocol when preparing nuclear extracts for Western blot.

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.

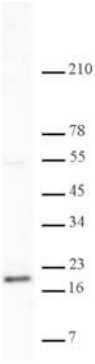
This product is for research use only and is not for use in diagnostic procedures.



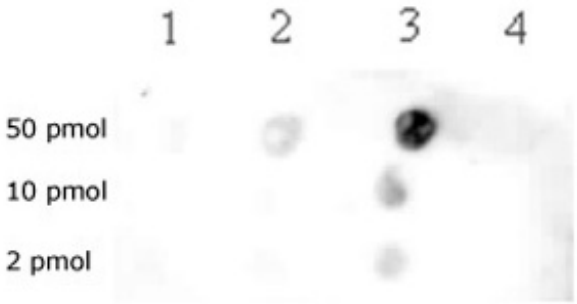
Histone H3K79me2 antibody (pAb) tested by ChIP-chip.
ChIP was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) with chromatin from 3 million HeLa cells and 5 µl of antibody. ChIP DNA was amplified by WGA, labeled and hybridized to a human tiling array. The images are characteristic of the binding patterns seen at many genes across the genome with the highest binding occurring at the promoter and a gradual decrease as signal moves into the gene body.



Histone H3 dimethyl Lys79 antibody (pAb) tested by ChIP analysis.
Chromatin IP performed using the ChIP-IT® Express Kit (Catalog No. 53008) and HeLa chromatin (1.5 x 10^6 cell equivalents per ChIP) using 10 µl of Histone H3 dimethyl Lys79 antibody or the equivalent amount of rabbit IgG as a negative control. Real time, quantitative PCR (RT-qPCR) was performed on ChIP DNA using a primer pair specific for the indicated gene. Data are presented as Fold Enrichment of the ChIP antibody signal compared to the negative control IgG (which has been normalized to 1.0) using the ddCT method.



Histone H3 dimethyl Lys79 antibody tested by Western blot.
HeLa acid extract (10 µg) was probed with Histone H3 dimethyl Lys79 antibody (1:2,500 dilution).



Histone H3 dimethyl Lys79 antibody tested by dot blot analysis.
Dot blot analysis was used to confirm the specificity of Histone H3 dimethyl Lys79 antibody for dimethyl lysine 79 of histone H3. Decreasing amounts of peptides corresponding to the region around lysine 79 of histone H3 were spotted onto PVDF and probed with the antibody at a 1:10,000 dilution.

Lane 1: Unmodified Lys79 peptide.
Lane 2: Monomethyl Lys79 peptide.
Lane 3: Dimethyl Lys79 peptide.
Lane 4: Trimethyl Lys79 peptide.