

# Histone H3K79me1 antibody (pAb)

Catalog Nos: 39921, 39922

RRID: AB\_2793394 Isotype: IgG Application(s): ChIP, DB, WB Reactivity: Human, Wide Range Predicted Quantities: 100 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). 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 monomethyl Lys79 antibody was raised against a peptide including monomethyl-lysine 79 of histone H3.

**Buffer:** Purified IgG in PBS (pH 7.5) with 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic. For your convenience, an unpurified serum version (Catalog No. 39145) of this antibody is also available.

## **Application Notes:**

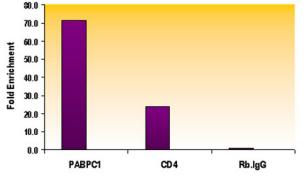
Applications Validated by Active Motif: ChIP: 10 µg per ChIP WB\*: 1 - 2 µg/ml dilution

\*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. The addition of 0.05% Tween 20 in the blocking buffer and primary antibody incubation buffer is recommended to aid in detection by Western blot. Individual optimization may be required.

**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 H3 monomethyl Lys79 antibody tested by ChIP analysis.

Chromatin IP performed using the ChIP-IT<sup>®</sup> Express Kit (Catalog No. 53008) and HeLa Chromatin ( $1.5 \times 10^6$  cell equivalents per ChIP) using 10 µg of Histone H3 monomethyl Lys79 pAb 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.

# $\begin{array}{c} - 195 \\ - 142 \\ - 96 \\ - 71 \\ - 48 \\ - 33 \\ - 28 \\ - 22 \\ - 12 \\ - 7.6 \\ \end{array}$

#### Histone H3 monomethyl Lys79 antibody (pAb) tested by Western blot.

Nuclear extract of HeLa cells (20  $\mu$ g) probed with Histone H3 monomethyl Lys79 antibody (1  $\mu$ g/ml dilution).

# 1 2 3 4 5 6 7 8 9 10 250 50 10

### Histone H3 monomethyl Lys79 antibody (pAb) tested by dot blot analysis.

Dot blot analysis was used to confirm the specificity of Histone H3 monomethyl Lys79 antibody for monomethyl-Lys79 of histone H3. Peptides corresponding to the region around lysine 79 of histone H3 were spotted onto PVDF and probed with Histone H3 monomethyl Lys79 antibody at a dilution of 1 µg/ml. The amount of peptide (in picomoles) spotted is indicated next to each row. Lane 1: unmodified Lys64 peptide. Lane 2: monomethyl Lys64 peptide. Lane 3: dimethyl Lys64 peptide. Lane 4: trimethyl Lys64 peptide. Lane 5: monomethyl Lys79 peptide. Lane 6: dimethyl Lys56 peptide. Lane 7: trimethyl Lys56 peptide. Lane 8: monomethyl Lys79 peptide. Lane 9: dimethyl Lys79 peptide. Lane 10: trimethyl Lys79 peptide. No detection of peptides (unmodified, mono-, di-, or tri-methylated) corresponding to lysine 4, lysine 9, lysine 18, lysine 23, lysine 27 and lysine 36 of Histone H3 was observed with Histone H3 dimethyl Lys79 antibody.