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## Data Sheet

### Anti-H3K9me3 monoclonal antibody

Catalog #: 25271

<b>Lot #:</b> 150415	<b>Host Species:</b> Mouse
<b>Conc.:</b> 1.8 µg/µl	<b>Species Reactivity:</b> Human
<b>Size:</b> 50 µg/28 µl	<b>Immunogen:</b> Synthetic peptide
<b>Clonality:</b> Monoclonal	<b>Purification:</b> Protein A purified

**Description:** Monoclonal antibody raised in Mouse against histone H3 trimethylated at lysine 9 (H3K9me3), using a KLH-conjugated synthetic peptide.

**Background:** Trimethylation of histone H3K9 is associated with satellite repeat regions and ZNF repeat genes, and correlates with gene repression.

**Formulation:** PBS containing 0.05% azide and 0.05% ProClin 300

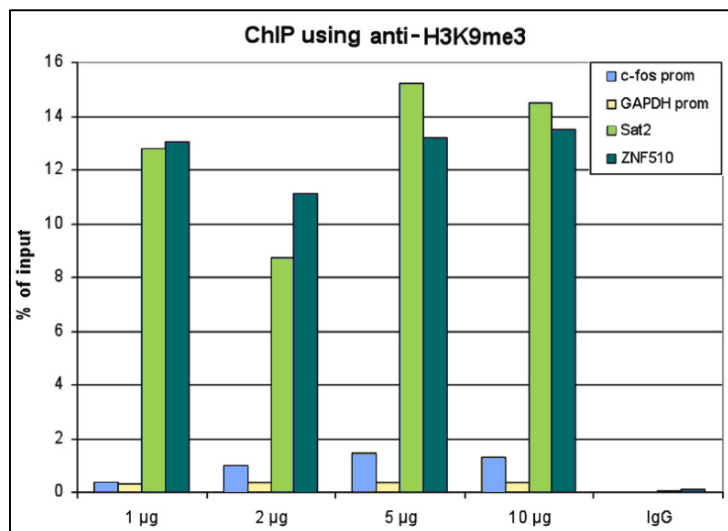
**Applications:** ChIP (1 µg/ChIP), DB (1:100,000), WB (1:1000)

**Storage/Stability:** Store at -80°C for up to 2 years. Centrifuge after first thaw to maximize product recovery. Aliquot to avoid repeated freeze/thaw cycles. Aliquots may be stored at -20°C for at least one month.

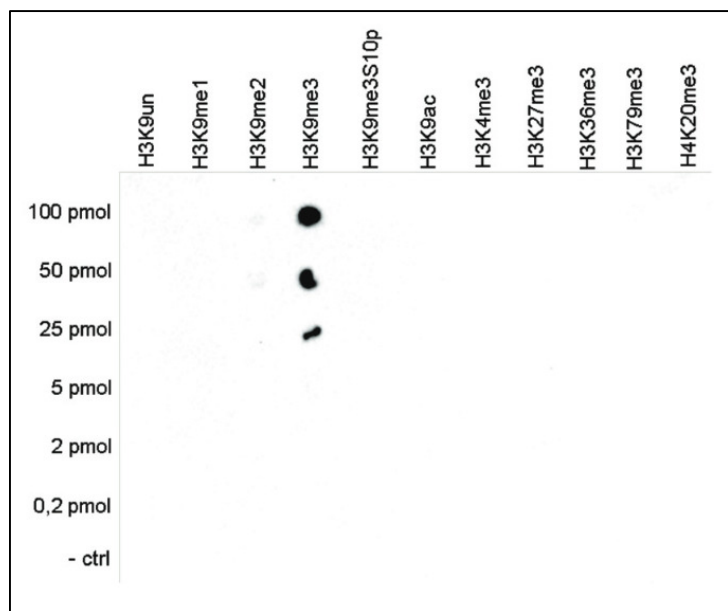
**Warnings:** Avoid freeze/thaw cycles

**Notes:** The optimal antibody amount per IP should be determined by the end-user. We recommend testing 1-5 µg per IP.

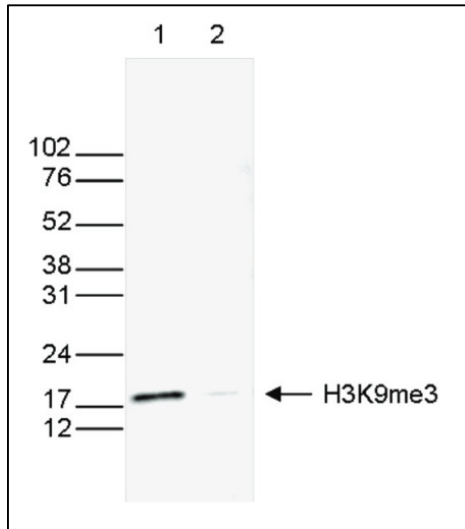
**Quality Assurance:**



ChIP results obtained with the monoclonal antibody directed against H3K9me3. ChIP assays were performed on human HeLa cells using the monoclonal antibody against H3K9me3 (cat. No. 25271). ChIP was performed using sheared chromatin from 1,000,000 cells. A titration consisting of 1, 2, 5 and 10 µg of antibody per ChIP experiment was analyzed. IgG (2 µg/IP) was used as a negative IP control. QPCR was performed with primers for the promoters of the active c-fos and GAPDH genes, used as negative controls, and for the ZNF510 gene and the Sat2 satellite repeat region, used as positive controls. Figure 1 shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).



Cross reactivity test using the monoclonal antibody directed against H3K9me3. A Dot Blot analysis was performed to test the cross reactivity of the monoclonal antibody against H3K9me3 (cat. No. 25271) with peptides containing different modifications of histone H3 or H4 and the unmodified H3K9 sequence. One hundred to 0.2 pmol of peptide containing the respective histone modification were spotted on a membrane. The antibody was used at a dilution of 1:100,000. Figure 2 shows a high specificity of the antibody for the modification of interest.



**Western blot analysis using the monoclonal antibody directed against H3K9me3.**

Western blot was performed on histone extracts (15 µg, lane 1) from HeLa cells, and on 1 µg of recombinant histone H3 (lane 2) using the monoclonal antibody against H3K9me3 (cat. No. 25271). The antibody was diluted 1:1,000 in TBS-Tween containing 5% skimmed milk. The marker (in kDa) is shown on the left, the position of the protein is indicated on the right.