

AbFlex® Histone H3K4me1 antibody (rAb)

Catalog Nos: 91415, 91416

Application(s): ChIP-Seq, CUT&Tag, WB

Reactivity: Human, Wide Range Predicted

Quantities: 100 µg, 10 µg

Purification: Protein A Chromatography

Host: Rabbit

Isotype: IgG

Concentration: 1 µg/µl

Molecular Weight: 17 kDa

Background:

AbFlex® antibodies are recombinant antibodies (rAbs) that have been generated using defined DNA sequences to produce highly specific, reproducible antibodies. Each AbFlex antibody contains a 6xHis Tag, a Biotinylation Tag for enzymatic biotin conjugation using the biotin ligase, BirA, and a sortase recognition motif (LPXTG) to attach a variety of labels directly to the antibody including fluorophores, enzymatic substrates (HRP, AP), peptides, drugs as well as solid supports. AbFlex® Histone H3K4me1 antibody was expressed as full-length IgG with mouse immunoglobulin heavy and light chains (IgG2a isotype) in mammalian 293 cells.

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). Histone H1 is a linker histone, present at the interface between the nucleosome core and DNA entry/exit points. Histone H1 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; these modifications play a major role in regulating gene expression.

Immunogen: This antibody was raised against a peptide containing mono-methyl lysine 4 of Histone H3.

Buffer: Purified IgG in 140 mM Hepes, pH 7.5, 70 mM NaCl, 32 mM NaOAc, 30% glycerol and 0.035% sodium azide. Sodium azide is highly toxic.

Application Notes:

Applications Validated by Active Motif:

ChIP-Seq: 5 µg each

WB*: 0.5 - 2 µg/ml dilution

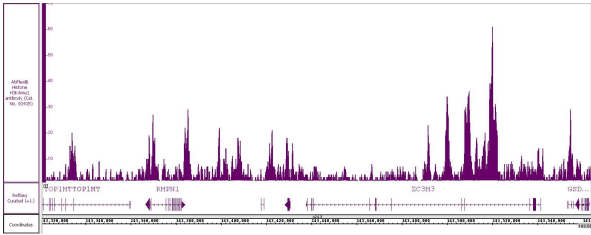
HPA: 1:20000 dilution.

CUT&Tag: 1-2 µg per 50 µl reaction

*For optimal results, primary antibody incubations should be performed at room temperature. The addition of 0.05% Tween 20 to all blocking solutions may also reduce background. Individual optimization may be required.

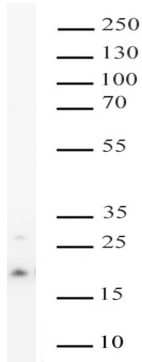
Storage and Guarantee: Some products may be shipped at room temperature. This will not affect their stability or performance. Avoid repeated freeze/thaw cycles by aliquoting items into single-use fractions for storage at -20°C for up to 2 years. Keep all reagents on ice when not in storage. This product is guaranteed for 12 months from date of receipt.

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



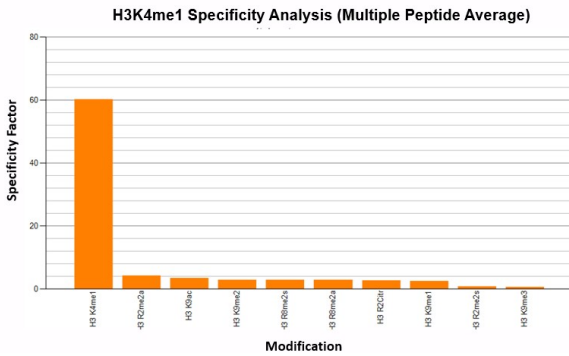
AbFlex® Histone H3K4me1 rAb antibody tested by ChIP-Seq.

ChIP was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) with 30 ug of chromatin from HeLa cells and 5 µg of antibody. ChIP DNA was sequenced on the Illumina HiSeq and 36 million sequence tags were mapped to identify H3K4me1 binding sites. The image shows binding across a region of chromosome 8.



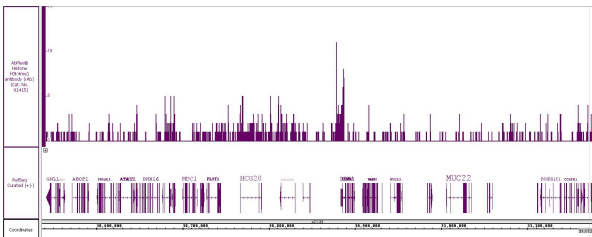
AbFlex® Histone H3K4me1 rAb antibody tested by Western blot.

Detection of H3K4me1 by Western blot analysis using 20 µg HeLa nuclear extract and probed with antibody at 0.5 µg/ml.



AbFlex® Histone H3K4me1 antibody (rAb) specificity tested by Peptide Array analysis.

Peptide array analysis was used to confirm the specificity of this antibody for its intended modification. AbFlex® Histone H3K4me1 antibody (rAb) was applied to Active Motif's MODified™ Histone Peptide Array (Catalog No. 13001) at a dilution of 1:20000. The arrays were scanned with ArrayAnalysis Software 7 and the results plotted.



AbFlex® Histone H3K4me1 antibody (rAb) tested by CUT&Tag

CUT&Tag was performed using 100,000 K562 cells and sequenced using 38 base-pair, paired-end reads on the Illumina NovaSeq. Data was collected from 4 million reads, and H3K4me1 data is shown for Chromosome 6.