

AbFlex[®] Histone H3K36me3 antibody (rAb)

Catalog Nos: 91265, 91266

RRID: AB_2793825 Isotype: IgG2a Application(s): ChIP, ChIP-Seq, WB Reactivity: Human, Wide Range Predicted Quantities: 100 µg, 10 µg Purification: Protein A Chromatography Host: Mouse 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[®] H3K36me3 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). 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. The methylation of histones can occur on two different residues: arginine or lysine. Histone H3 is methylated at lysine 36 by the Set2 (yeast) and NSD1 (mammals) methyltransferases. Dimethylation of lysine 36 of histone H3 is involved with transcriptional elongation by RNA pol II holoenzyme and is a marker of transcribed genes.

Immunogen: This antibody was raised against a peptide containing trimethly-lysine 36 of histone H3.

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

Application Notes:

Applications Validated by Active Motif: ChIP-Seq: 4 µg per ChIP WB*: 0.5 - 2 µg/ml

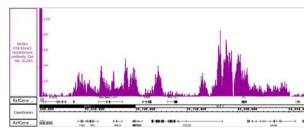
*Note: Many chromatin-bound proteins are not soluble in a low salt nuclear extract and fractionate to the pellet. Therefore, we recommend a High Salt / Sonication Protocol when preparing nuclear extracts for Western blot.

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





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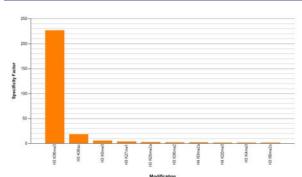
= 28 22 = 12 = 7.6

AbFlex® Histone H3K36me3 recombinant antibody (rAb) tested by ChIP-Seq

Chromatin immunoprecipitation (ChIP) was performed using the ChIP-IT® High Sensitivity Kit (Cat. No. 53040) with 30 μ g of HAP1 cell line- myeloid leukemia chromatin and 4 μ g of Histone H3K36me3 antibody. ChIP DNA was sequenced on the Illumina NextSeq and 15.4 million sequence tags were mapped to identify Histone H3K36me3 binding sites on chromosome 16.

AbFlex® Histone H3K36me3 recombinant antibody (rAb) tested by Western blot

20 μ g of HeLa cell nuclear extract was run on SDS-PAGE and probed with AbFlex H3K36me3 antibody at 2 μ g/ml.



Histone H3K36me3 antibody (rAb) specificity tested by peptide array analysis.

Peptide array analysis was used to confirm the specificity of this antibody for its intended modification. Histone H3K36me3 antibody was applied at a dilution of 0.2 µg/ml to Active Motif's MODified[™] Histone Peptide Array (Catalog No. 13001). The arrays were scanned with ArrayAnalysis Software 16 and the results plotted. Specificity data is shown for the most reactive peptides and those related to the immunogen.