# abcam

## Product datasheet

# Anti-Histone H3 (di methyl K36) antibody - ChIP Grade ab9049

★★★★★ 22 Abreviews 123 References 4 图像

概述

产品名称 Anti-Histone H3 (di methyl K36)抗体- ChIP Grade

描述 兔多克隆抗体to Histone H3 (di methyl K36) - ChIP Grade

**宿主** Rabbit

特异性 This antibody is specific for histone H3 di-methylated at K36. It does not react with mono or tri

methyl K36.

经测试应用 适用于: IHC-P, WB, ChIP

种属反应性 与反应: Cow, Human, Drosophila melanogaster

预测可用于: Mouse, Pig, Saccharomyces cerevisiae, Arabidopsis thaliana, Caenorhabditis

elegans, Zebrafish, Mammals, Dictyostelium discoideum

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

(Peptide available as ab1784)

常规说明

The Life Science industry has been in the grips of a reproducibility crisis for a number of years.

Abcam is leading the way in addressing this with our range of recombinant monoclonal antibodies and knockout edited cell lines for gold-standard validation. Please check that this product meets

your needs before purchasing.

If you have any questions, special requirements or concerns, please send us an inquiry and/or contact our Support team ahead of purchase. Recommended alternatives for this product can be

found below, along with publications, customer reviews and Q&As

性能

形式 Liquid

**存放说明** Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

**存储溶液** pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

 $agent. \ \ \ If you would \ like information about the formulation of a specific lot, please contact our$ 

scientific support team who will be happy to help.

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纯**度** Immunogen affinity purified

应用

#### The Abpromise guarantee Abpromise™承诺保证使用ab9049于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应 <b>用</b>	Ab评论	说明
IHC-P	★★★★★ (2)	Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
WB	★★★★★ (14)	Use a concentration of 1 µg/ml. Detects a band of approximately 17 kDa (predicted molecular weight: 15 kDa).Can be blocked with Human Histone H3 (di methyl K36) peptide (ab1784).
ChIP	****(3)	Use 2-5 μg for 25 μg of chromatin. Use RPL30 ChIP primer pair <u>ab269262</u> as positive control.

#### 靶标

功能

Core component of nucleosome. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility is regulated via a complex set of post-translational modifications of histones, also called histone code, and nucleosome remodeling.

序列相似性

发展阶段

Belongs to the histone H3 family.

翻译后修饰

Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

Acetylation is generally linked to gene activation. Acetylation on Lys-10 (H3K9ac) impairs methylation at Arg-9 (H3R8me2s). Acetylation on Lys-19 (H3K18ac) and Lys-24 (H3K24ac) favors methylation at Arg-18 (H3R17me).

Citrullination at Arg-9 (H3R8ci) and/or Arg-18 (H3R17ci) by PAD4 impairs methylation and represses transcription.

Asymmetric dimethylation at Arg-18 (H3R17me2a) by CARM1 is linked to gene activation. Symmetric dimethylation at Arg-9 (H3R8me2s) by PRMT5 is linked to gene repression.

Asymmetric dimethylation at Arg-3 (H3R2me2a) by PRMT6 is linked to gene repression and is mutually exclusive with H3 Lys-5 methylation (H3K4me2 and H3K4me3). H3R2me2a is present at the 3' of genes regardless of their transcription state and is enriched on inactive promoters, while it is absent on active promoters.

Methylation at Lys-5 (H3K4me), Lys-37 (H3K36me) and Lys-80 (H3K79me) are linked to gene activation. Methylation at Lys-5 (H3K4me) facilitates subsequent acetylation of H3 and H4. Methylation at Lys-80 (H3K79me) is associated with DNA double-strand break (DSB) responses and is a specific target for TP53BP1. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are linked to gene repression. Methylation at Lys-10 (H3K9me) is a specific target for HP1

proteins (CBX1, CBX3 and CBX5) and prevents subsequent phosphorylation at Ser-11 (H3S10ph) and acetylation of H3 and H4. Methylation at Lys-5 (H3K4me) and Lys-80 (H3K79me) require preliminary monoubiquitination of H2B at 'Lys-120'. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are enriched in inactive X chromosome chromatin.

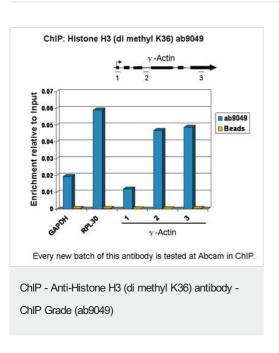
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and RPS6KA5 is important during interphase because it enables the transcription of genes following external stimulation, like mitogens, stress, growth factors or UV irradiation and result in the activation of genes, such as c-fos and c-jun. Phosphorylation at Ser-11 (H3S10ph), which is linked to gene activation, prevents methylation at Lys-10 (H3K9me) but facilitates acetylation of H3 and H4. Phosphorylation at Ser-11 (H3S10ph) by AURKB mediates the dissociation of HP1 proteins (CBX1, CBX3 and CBX5) from heterochromatin. Phosphorylation at Ser-11 (H3S10ph) is also an essential regulatory mechanism for neoplastic cell transformation. Phosphorylated at Ser-29 (H3S28ph) by MLTK isoform 1, RPS6KA5 or AURKB during mitosis or upon ultraviolet B irradiation. Phosphorylation at Thr-7 (H3T6ph) by PRKCBB is a specific tag for epigenetic transcriptional activation that prevents demethylation of Lys-5 (H3K4me) by LSD1/KDM1A. At centromeres, specifically phosphorylated at Thr-12 (H3T11ph) from prophase to early anaphase, by DAPK3 and PKN1. Phosphorylation at Thr-12 (H3T11ph) by PKN1 is a specific tag for epigenetic transcriptional activation that promotes demethylation of Lys-10 (H3K9me) by KDM4C/JMJD2C. Phosphorylation at Tyr-42 (H3Y41ph) by JAK2 promotes exclusion of CBX5 (HP1 alpha) from chromatin.

Monoubiquitinated by RAG1 in lymphoid cells, monoubiquitination is required for V(D)J recombination (By similarity). Ubiquitinated by the CUL4-DDB-RBX1 complex in response to ultraviolet irradiation. This may weaken the interaction between histones and DNA and facilitate DNA accessibility to repair proteins.

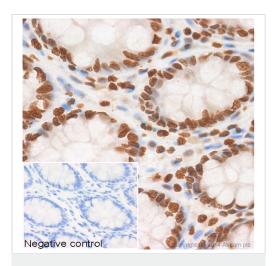
细胞定位

Nucleus. Chromosome.

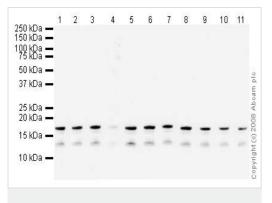
#### 图片



Chromatin was prepared from Hela cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 min. The ChIP was performed with 25  $\mu$ g of chromatin, 2  $\mu$ g of ab9049(blue), and 20  $\mu$ l of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified on the GAPDH and RPL30 (active) promoters and over the g-Actin gene (active). Schematic diagram of the g-Actin gene is shown on the top of the figure. Black boxes represent exons and thin lines represent introns. PCR products are depicted as bars under the gene.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049)



Western blot - Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049)

IHC image of ab9049 staining Histone H3 (di methyl K36) in human colon formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab9049, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the negative control (shown on the inset).

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

**All lanes :** Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049) at 1  $\mu$ g/ml

Lane 1 : Calf Thymus Histone Preparation Nuclear Lysate

Lane 2 : Calf Thymus Histone Preparation Nuclear Lysate with

Histone H3 peptide (ab41430) at 0.5 µg/ml

**Lane 3 :** Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K36) peptide (ab1783) at 0.5  $\mu g/ml$ 

**Lane 4 :** Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K36) peptide (<u>ab1784</u>) at 0.5 µg/ml

**Lane 5**: Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K36) peptide (ab1785) at 0.5 µg/ml

**Lane 6 :** Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K9) peptide (<u>ab1771</u>) at 0.5 μg/ml

Lane 7: Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K9) peptide (ab1772) at 0.5 µg/ml

**Lane 8 :** Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K9) peptide (**ab1773**) at 0.5 µg/ml

**Lane 9 :** Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K27) peptide (ab1780) at 0.5  $\mu$ g/ml

**Lane 10**: Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K27) peptide (<u>ab1781</u>) at 0.5 μg/ml **Lane 11**: Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K27) peptide (<u>ab1782</u>) at 0.5 μg/ml

Lysates/proteins at 0.5 µg per lane.

#### Secondary

**All lanes :** Goat polyclonal to Rabbit lgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 15 kDa Observed band size: 17 kDa

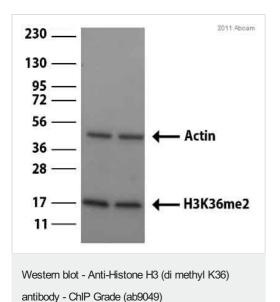


Image courtesy of an anonymous Abreview.

**All lanes :** Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049) at 1/1400 dilution

**All lanes :** Whole cell lysate prepared from Drosophila BG3 tissue cells

Lysates/proteins at 50000 cells per lane.

#### **Secondary**

**All lanes :** Donkey anti-rabbit polyclonal conjugated to HRP at 1/20000 dilution

Developed using the ECL technique.

**Predicted band size:** 15 kDa **Observed band size:** 17,42 kDa

Exposure time: 30 seconds

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