

Product datasheet

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

★★★★★ 20 Abreviews 61 References 6 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, ICC/IF, WB, ChIP
种属反应性	与反应: Mouse, Cow, Human, Pig, Saccharomyces cerevisiae, Arabidopsis thaliana, Caenorhabditis elegans, Drosophila melanogaster, Zebrafish, Dictyostelium discoideum 预测可用于: Mammals
免疫原	Synthetic peptide within Human Histone H3 aa 1-100 (di methyl K36) conjugated to Keyhole Limpet Haemocyanin (KLH). The exact sequence is proprietary. (Peptide available as ab1784)
阳性对照	WB: Calf thymus histone preparation and HeLa whole cell extract. IHC-P: Human normal colon tissue.

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
纯度	Immunogen affinity purified
克隆	多克隆
同种型	IgG

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.

应用

Our [Abpromise guarantee](#) covers the use of **ab9049** in the following tested applications.

The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

应用	Ab评论	说明
IHC-P	★★★★★	Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF	★★★★★	1/200.
WB	★★★★★	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	★★★★★	Use 2-5 µg for 25 µg of chromatin.

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	Belongs to the histone H3 family.
发展阶段	Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.
翻译后修饰	<p>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).</p> <p>Citrullination at Arg-9 (H3R8ci) and/or Arg-18 (H3R17ci) by PADI4 impairs methylation and represses transcription.</p> <p>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.</p> <p>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.</p> <p>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</p>

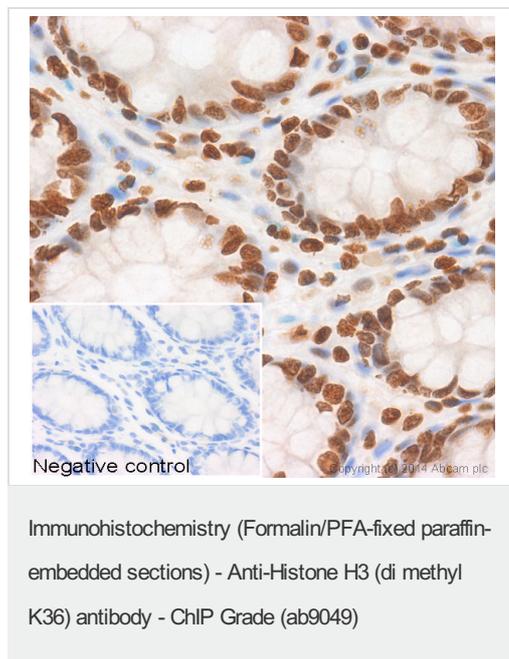
at Ser-11 (H3S10ph) by RPS6KA4 and RPS6KA5 is important during interphase because it enables the transcription of genes following external stimulation, like mitogens, stress, growth factors or UV irradiation and result in the activation of genes, such as c-fos and c-jun. Phosphorylation at Ser-11 (H3S10ph), 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

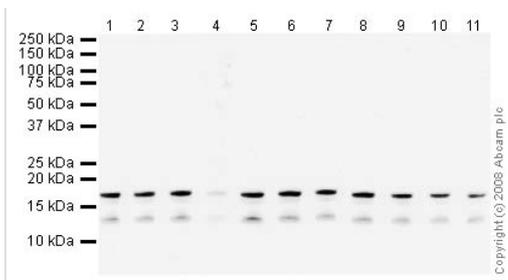
Nucleus. Chromosome.

图片



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.



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

All lanes : Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049) at 1 µg/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 µg/ml

Lane 4 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K36) peptide (ab1784) 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 (ab1771) 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 µg/ml

Lane 10 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K27) peptide (ab1781) at 0.5 µg/ml

Lane 11 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K27) peptide (ab1782) at 0.5 µg/ml

Lysates/proteins at 0.5 µg per lane.

Secondary

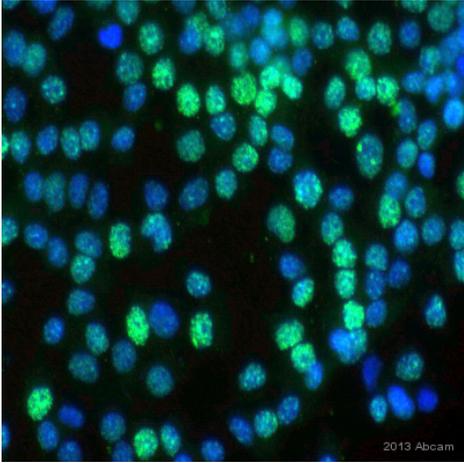
All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/3000 dilution

Performed under reducing conditions.

Predicted band size: 15 kDa

Observed band size: 17 kDa

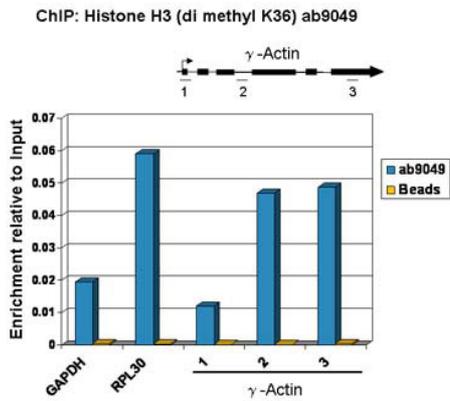
[why is the actual band size different from the predicted?](#)



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049)

This image is courtesy of an anonymous Abreview

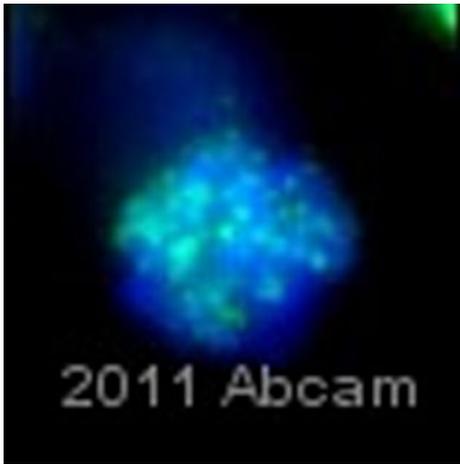
ab9049 staining Histone H3 (di methyl K36) in Mouse Neuro-2a neuroblastoma cultured cells by ICC/IF (Immunocytochemistry/immunofluorescence). Cells were fixed with formaldehyde, permeabilized with 0.3% Triton X-100 and blocked with 1% BSA for 1 hour at 25°C. Samples were incubated with primary antibody (1/100) for 16 hours at 4°C. A FITC-conjugated Goat anti-rabbit IgG polyclonal (1/1000) was used as the secondary antibody.



Every new batch of this antibody is tested at Abcam in ChIP.

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

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 µg of chromatin, 2 µg of ab9049 (blue), and 20 µl 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.



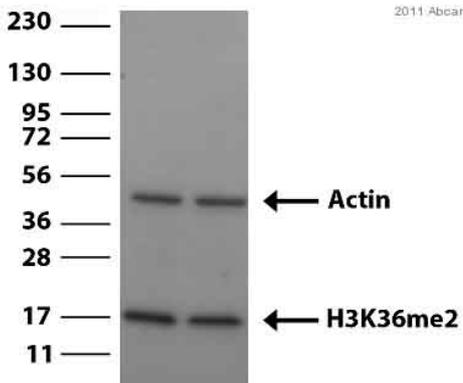
Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K36) antibody - ChIP Grade (ab9049)

Image courtesy of an anonymous Abreview.

ab9049 staining Histone H3 (di methyl K36) in C elegans intestinal cells by Immunocytochemistry/ Immunofluorescence.

Cells were fixed in paraformaldehyde and permeabilized using sperm salts buffer, Triton X-100. Samples were then incubated with ab9049 at a 1/200 for 16 hours at 22°C. A FITC conjugated goat polyclonal was used as the secondary at a 1/100 dilution.

Counterstained with DAPI.



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

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 [why is the actual band size different from the predicted?](#)

Exposure time: 30 seconds

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