

Product datasheet

Anti-Histone H3 (phospho T11) antibody - ChIP Grade ab5168

★★★★☆ 3 Abreviews 10 References 5 图像

概述

产品名称	Anti-Histone H3 (phospho T11)抗体- ChIP Grade
描述	兔多克隆抗体to Histone H3 (phospho T11) - ChIP Grade
宿主	Rabbit
经测试应用	适用于: WB, ChIP, IHC-P, ICC/IF
种属反应性	与反应: Mouse, Cow, Human 预测可用于: Rat, Saccharomyces cerevisiae, Xenopus laevis, Arabidopsis thaliana, Caenorhabditis elegans, Drosophila melanogaster, Schizosaccharomyces pombe, Zebrafish, Neurospora crassa
免疫原	Synthetic peptide corresponding to Human Histone H3 aa 1-100 (phospho T11) conjugated to Keyhole Limpet Haemocyanin (KLH). Database link: P68431 (Peptide available as ab24444)
阳性对照	Calf thymus histone lysate; Colcemid treated HeLa Histone prep; HeLa (Human epithelial carcinoma cell line) Nuclear Lysate IHC-P: human normal testis FFPE tissue sections

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	Preservative: 0.02% Sodium Azide Constituents: 1% BSA, PBS. pH 7.4
纯度	Immunogen affinity purified
克隆	多克隆
同种型	IgG

应用

Our [Abpromise guarantee](#) covers the use of **ab5168** 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
WB	★★★★☆	1/500 - 1/1000. Detects a band of approximately 15 kDa (predicted molecular weight: 15 kDa).
ChIP		Use at an assay dependent concentration.
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		Use a concentration of 1 µg/ml.

靶标

功能

Variant histone H3 which replaces conventional H3 in a wide range of nucleosomes in active genes. Constitutes the predominant form of histone H3 in non-dividing cells and is incorporated into chromatin independently of DNA synthesis. Deposited at sites of nucleosomal displacement throughout transcribed genes, suggesting that it represents an epigenetic imprint of transcriptionally active chromatin. 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 throughout the cell cycle independently of DNA synthesis.

翻译后修饰

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.

Specifically enriched in modifications associated with active chromatin such as methylation at Lys-5 (H3K4me), Lys-37 and Lys-80. 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), which are linked to gene repression, are underrepresented. 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.

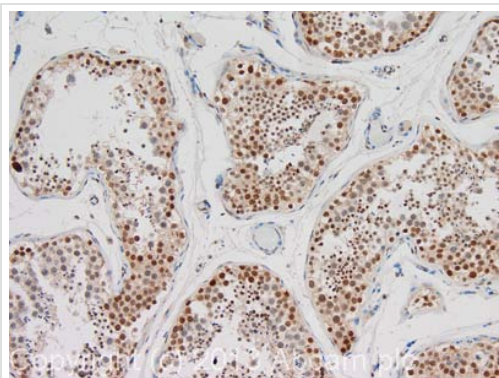
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and 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. Phosphorylation on Ser-32 (H3S31ph) is specific to regions bordering centromeres in metaphase chromosomes. Ubiquitinated. Monoubiquitinated by RAG1 in lymphoid cells, monoubiquitination is required for V(D)J recombination.

细胞定位

Nucleus. Chromosome.

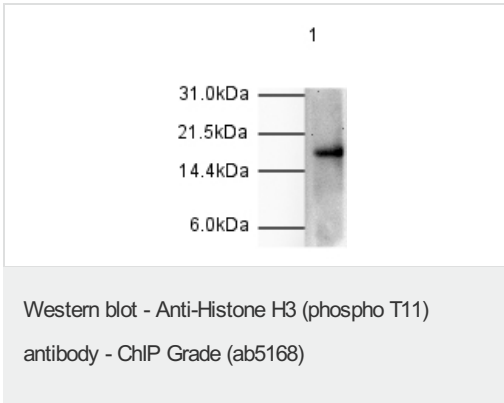
图片



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (phospho T11) antibody - ChIP Grade (ab5168)

IHC image of Histone H3 (phospho T11) staining in human normal testis formalin fixed paraffin embedded tissue section, performed on a Leica Bond™ system using the standard protocol F. 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 ab5168, 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.

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.



Anti-Histone H3 (phospho T11) antibody - ChIP Grade (ab5168) at 1/500 dilution + Calf thymus histone lysate

Secondary

Goat Anti-Rabbit IgG H&L (HRP) (ab6721) at 1/2000 dilution

Performed under reducing conditions.

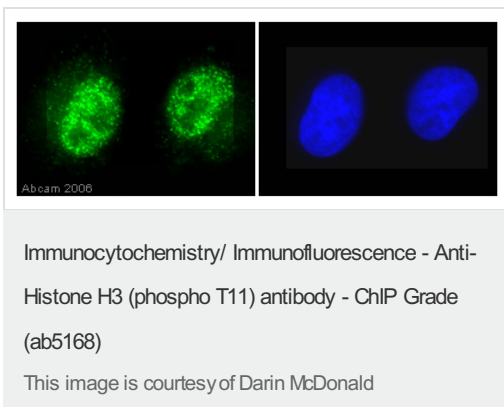
Predicted band size: 15 kDa

Observed band size: 17 kDa

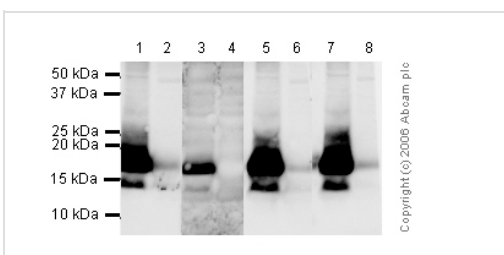
Rabbit polyclonal to Histone H3 (phospho T11) - ab5168 at 1/500 dilution.

Lane 1 contains 1ug calf thymus histone prep.

Secondary ab: Goat anti-rabbit IgG HRP conjugate ab6721 (1/2000)



SKN-SH cells were fixed in 4% paraformaldehyde for 10 mins, permeabilized in PBS-0.5% Triton X-100 for 5 mins and incubated for 30 minutes with ab5168 (1/100). The slides were rinsed once in PBS-Triton (0.1%), twice in PBS then incubated with the secondary antibody for 30 mins. The DNA is stained with DAPI (blue). Clear nuclear staining with ab5168 can be seen (green). 100x magnification.



All lanes : Anti-Histone H3 (phospho T11) antibody - ChIP Grade (ab5168) at 1 µg/ml

Lane 1 : Colcemid treated HeLa Histone prep at 5 µg

Lane 2 : HeLa (Human epithelial carcinoma)

Western blot - Anti-Histone H3 (phospho T11)
antibody - ChIP Grade (ab5168)

cell line) Nuclear Lysate ([ab27251](#)) at 20 µg

Lane 3 : Colcemid treated HeLa Histone prep at 5 µg with Human Histone H3 (phospho T11) peptide ([ab24444](#)) at 1 µg/ml

Lane 4 : HeLa (Human epithelial carcinoma cell line) Nuclear Lysate ([ab27251](#)) at 20 µg with Human Histone H3 (phospho T11) peptide ([ab24444](#)) at 1 µg/ml

Lane 5 : Colcemid treated HeLa Histone prep at 5 µg with Human Histone H3 (unmodified) peptide ([ab2903](#)) at 1 µg/ml

Lane 6 : HeLa (Human epithelial carcinoma cell line) Nuclear Lysate ([ab27251](#)) at 20 µg with Human Histone H3 (unmodified) peptide ([ab2903](#)) at 1 µg/ml

Lane 7 : Colcemid treated HeLa Histone prep at 5 µg with Human Histone H3 (phospho S10) peptide ([ab11477](#)) at 1 µg/ml

Lane 8 : HeLa (Human epithelial carcinoma cell line) Nuclear Lysate ([ab27251](#)) at 20 µg with Human Histone H3 (phospho S10) peptide ([ab11477](#)) at 1 µg/ml

Secondary

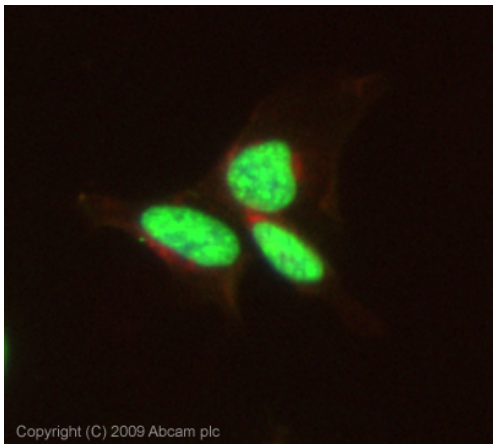
All lanes : Goat polyclonal to Rabbit IgG H&L (HRP) Pre-Adsorbed at 1/10000 dilution

Performed under reducing conditions.

Predicted band size: 15 kDa

Observed band size: 17 kDa

This antibody is blocked by the Histone H3 phospho T11 peptide but not by the respective unmodified peptide or the Histone H3 phospho S10 peptide. This confirms that the antibody is specific for phospho T11 of Histone H3.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (phospho T11) antibody - ChIP Grade (ab5168)

ICC/IF image of ab5168 stained Hek293 cells. The cells were 4% PFA fixed (10 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab5168, 1µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM. This antibody also gave a positive result in 4% PFA fixed (10 min) HeLa and HepG2 cells at 1µg/ml, and in 100% methanol fixed (5 min) HeLa, Hek293, HepG2 and MCF7 cells at 1µg/ml.

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