abcam

Product datasheet

Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade ab8895

58 Abreviews 1044 References 6 图像

概述

产品名称	Anti-Histone H3 (mono methyl K4)抗体- ChIP Grade
描述	兔多克隆抗体to Histone H3 (mono methyl K4) - ChIP Grade
宿主	Rabbit
特异性	Specific for mono-methylated Lysine 4 of histone H3. Does not recognise di- or tri-methyl Lysine 4 nor methylation at Lysine 9.
经测试应用	**适用于:** ICC/IF, ChIP, WB, IHC-P
种属反应性	**与反应:** Mouse, Rat, Cow, Human **预测可用于:** Pig, Saccharomyces cerevisiae, Tetrahymena, Xenopus laevis, Drosophila melanogaster, Plants, Mammals, Plasmodium falciparum, Xenopus tropicalis, Candida albicans
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab1340**)
阳性对照	ChIP: Chromatin prepared from U-2 OS cells; Metastatic clear cell renal carcinoma cells (M1A). WB: Calf thymus histone; HeLa; NIH3T3; PC12 ICC/IF: HeLa cells. IHC-P: Human colon tissue.
常规说明	Learn about ChIP assay kits, other ChIP antibodies, protocols and more in the **ChIP assay guide**. 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.
纯度	Immunogen affinity purified
克隆	**多克隆**
同种型	IgG

应用

The Abpromise guarantee **Abpromise™承诺保证使用ab8895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。

应用	Ab评论	说明
ICC/IF	★★★★★ (6)	Use a concentration of 1 - 5 µg/ml. Works better if cells fixed in methanol.
ChIP	★★★★☆ (16)	Use 2 µg for 25 µg of chromatin. We recommend Myo-D ChIP primer pair **ab269261** as positive control.
WB	★★★★☆ (22)	1/500. Detects a band of approximately 17 kDa (predicted molecular weight: 15 kDa).
IHC-P	★★★★☆ (4)	Use a concentration of 0.5 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

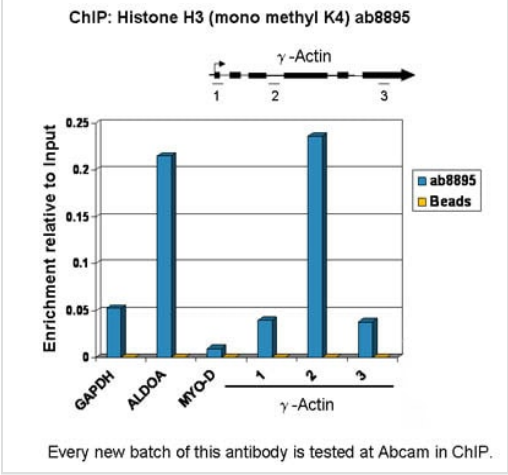
靶标

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	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 PADI4 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.

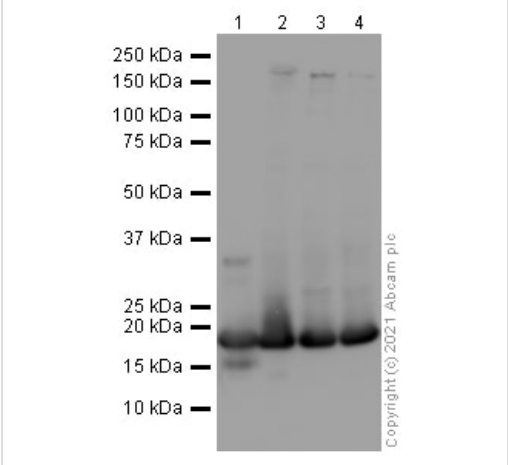
细胞定位 Nucleus. Chromosome.

图片



ChIP - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

Chromatin was prepared from U-2 OS cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 2µg of ab8895 (blue), and 20µl of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified on the GAPDH and ALDOA (active) and MYO-D (inactive) promoters and over the y-Actin gene (active). Schematic diagram of the y-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.



Western blot - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

All lanes : Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895) at 1 µg/ml

Lane 1 : Calf Thymus Histone at 0.5 µg
Lane 2 : HeLa Nuclear – Triton Prep at 10 µg
Lane 3 : NIH3T3 Nuclear – Triton Prep at 10 µg
Lane 4 : PC12 Nuclear at 10 µg

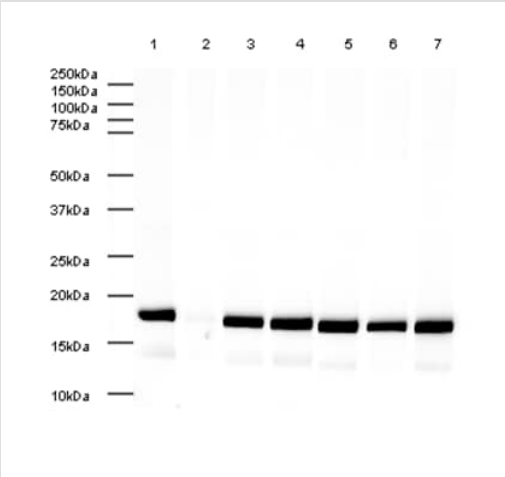
Secondary
All lanes : Goat polyclonal to Rabbit IgG - H&L - Pre-Adsorbed (HRP) at 1/50000 dilution

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

Exposure time: 30 seconds

Blocking buffer: 2% BSA

Gel type: MES



Western blot - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

All lanes : Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895) at 1/500 dilution

Lane 1 : Calf thymus histone lysate
Lane 2 : Calf thymus histone lysate with Human Histone H3 (mono methyl K4) peptide (**ab1340**) at 1 µg/ml
Lane 3 : Calf thymus histone lysate with Human Histone H3 (di methyl K4) peptide (**ab7768**) at 1 µg/ml
Lane 4 : Calf thymus histone lysate with Human Histone H3 (tri methyl K4) peptide (**ab1342**) at 1 µg/ml
Lane 5 : Calf thymus histone lysate with Human Histone H3 (mono methyl K9) peptide (**ab1771**) at 1 µg/ml
Lane 6 : Calf thymus histone lysate with Human Histone H3 (mono methyl K27) peptide (**ab1780**) at 1 µg/ml
Lane 7 : Calf thymus histone lysate with Human Histone H3 (unmodified) peptide (**ab2903**) at 1 µg/ml

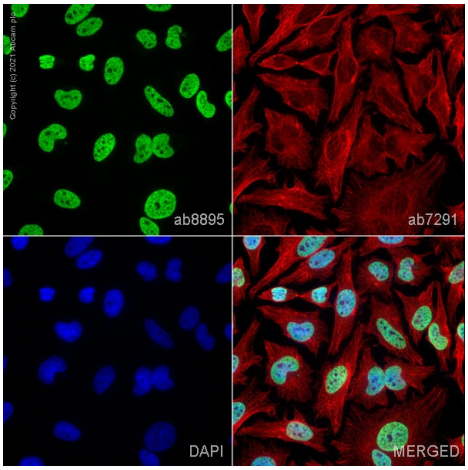
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab6721**) at 1/5000 dilution

Performed under reducing conditions.

Predicted band size: 15 kDa
Observed band size: 18 kDa

Exposure time: 2 minutes

ab8895 is specific for mono-methylated Lysine 4 of histone H3 and does not recognize di- or tri-methyl Lysine 4 nor methylation at Lysine 9. This is shown in lane 2 where the activity of the antibody is specifically blocked by the addition of the immunizing peptide (**ab1340**).

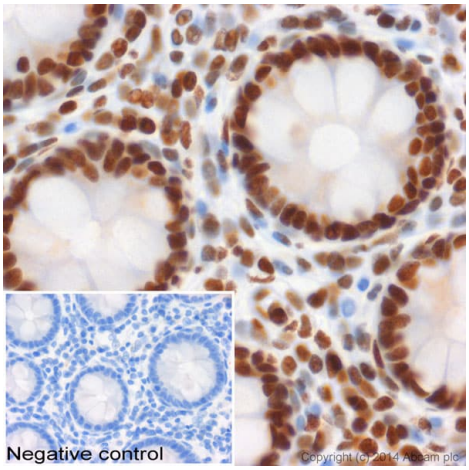


Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

ab8895 staining Histone H3 (mono methyl K4) in HeLa cells. The cells were fixed with 4% paraformaldehyde (10 min), permeabilized with 0.1% PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated overnight at 4°C with ab8895 at 1 µg/ml and **ab7291**, Mouse monoclonal [DM1A] to alpha Tubulin - Loading Control. Cells were then incubated with **ab150081**, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488), pre-adsorbed at 1/1000 dilution (shown in green) and **ab150120**, Goat polyclonal Secondary Antibody to Mouse IgG - H&L (Alexa Fluor® 594), pre-adsorbed at 1/1000 dilution (shown in pseudocolour red). Nuclear DNA was labelled with DAPI (shown in blue).

Also suitable in cells fixed with 100% methanol (5 min).

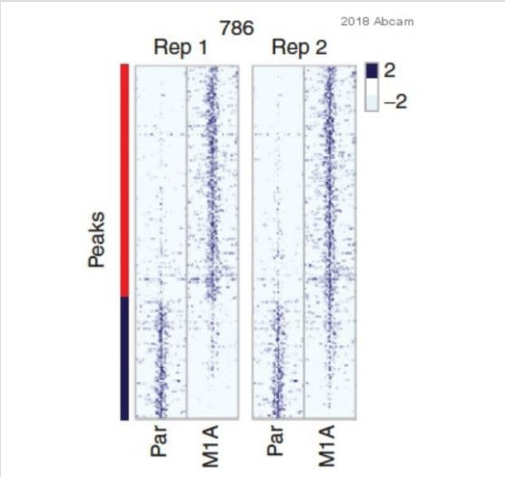
Image was acquired with a high-content analyser (Operetta CLS, Perkin Elmer) and a maximum intensity projection of confocal sections is shown.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

IHC image of ab8895 staining Histone H3 (mono methyl K4) 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 ab8895, 0.5µg/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.



ChIP - Anti-Histone H3 (mono methyl K4) antibody - ChIP Grade (ab8895)

This image is courtesy of an Abreview submitted by Paulo Rodrigues

ChIP analysis using ab8895 binding Histone H3 (mono methyl K4) in Metastatic clear cell renal carcinoma cells (M1A). Cells were cross-linked for 10 minutes with 1% formaldehyde. Samples were incubated at 1/100 dilution with primary antibody for 12 hours at 4°C.
Positive control:Metastatic clear cell renal carcinoma cells (M1A).
Negative Control:Parental clear cell renal carcinoma cells (786-O).

Our Abpromise to you: Quality guaranteed and expert technical support

- Replacement or refund for products not performing as stated on the datasheet
- Valid for 12 months from date of delivery
- Response to your inquiry within 24 hours
- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

If the product does not perform as described on this datasheet, we will offer a refund or replacement. For full details of the Abpromise, please visit **https://www.abcam.cn/abpromise** or contact our technical team.

Terms and conditions

- Guarantee only valid for products bought direct from Abcam or one of our authorized distributors