abcam

Product datasheet

Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade ab8898

★★★★★ 91 Abreviews 1603 References 画像数 5

製品の概要

製品名	Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade
製品の詳細	Rabbit polyclonal to Histone H3 (tri methyl K9) - ChIP Grade
由来種	Rabbit
特異性	Histone H3 (tri methyl K9) antibody (ab8898) is specific for Histone H3 tri methyl Lysine 9. Shows slight cross-reactivity with tri methyl K27, which shares a similar epitope (please see Western blot image). Does not react with mono or di methylated K9. From Jan 2024, QC testing of replenishment batches of this polyclonal changed. All tested and expected application and reactive species combinations are still covered by our Abcam product promise. However, we no longer test all applications. For more information on a specific batch, please contact our Scientific Support who will be happy to help. You may also be interested in our alternative recombinant antibody, **ab176916**.
アプリケーション	**適用あり:** WB, IHC-P, ChIP, ICC/IF
種交差性	**交差種:** Mouse, Cow, Human **交差が予測される動物種:** Rat, Chicken, Saccharomyces cerevisiae, Xenopus laevis, Drosophila melanogaster, Indian muntjac, Mammals, Xenopus tropicalis, Cyanidioschyzon merolae
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab1773**)
ポジティブ・コントロール	ChIP: U2OS cells, mouse ES cells. WB: Calf Thymus Histone Preparation Nuclear Lysate. IHC-P: Normal human colon. ICC: Mouse 3T3MEF, Indian muntjac fibroblast cells, HeLa cells, Mouse Embryonic Stem cells.
特記事項	Every new batch of ab8898 is tested in house in ChIP. 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

製品の特性

製品の状態	Liquid
保存方法	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.
バッファー	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.
精製度	Immunogen affinity purified
ポリ/モノ	ポリクローナル
アイソタイプ	IgG

アプリケーション

The Abpromise guarantee **Abpromise保証は、** 次のテスト済みアプリケーションにおけるab8898の使用に適用されます

アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
WB	★★★★★ (30)	Use at an assay dependent concentration. Predicted molecular weight: 15 kDa.Can be blocked with **Human Histone H3 (tri methyl K9) peptide (ab1773)**.
IHC-P	★★★★★ (9)	1/400. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ChIP	★★★★★ (25)	Use 2-4 µg for 25 µg of chromatin. We recommend SAT-alpha ChIP primer pair **ab269263** as a positive control.
ICC/IF	★★★★★ (21)	Use a concentration of 0.5 µg/ml.

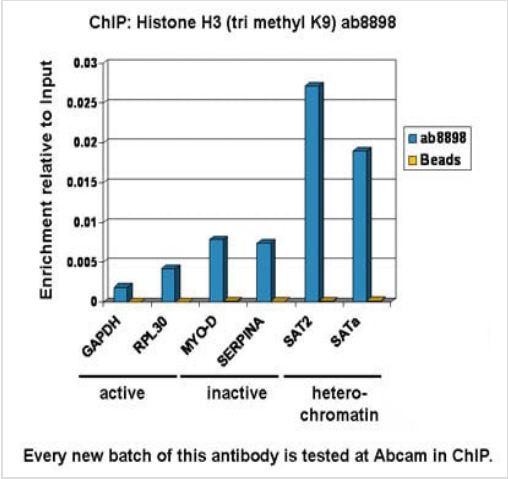
ターゲット情報

機能	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.
配列類似性	Belongs to the histone H3 family.
発生段階	Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.
翻訳後修飾	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.

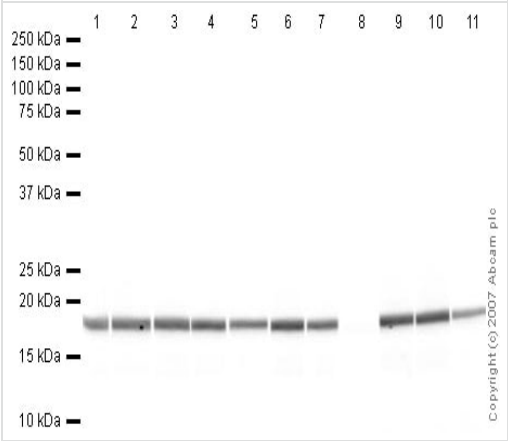
細胞内局在 Nucleus. Chromosome.

画像



ChIP - Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898)

Chromatin was prepared from U2OS 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 ab8898 (blue), and 20 µl of protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci). Primers and probes are located in the first kb of the transcribed region.



Western blot - Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898)

All lanes : Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898) at 1 µg/ml

Lane 1 : Calf Thymus Histone Preparation Nuclear Lysate
Lane 2 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (unmodified) peptide (**ab7228**) at 0.5 µg/ml
Lane 3 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (mono methyl K4) peptide (**ab1340**) at 0.5 µg/ml
Lane 4 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (di methyl K4) peptide (**ab7768**) at 0.5 µg/ml
Lane 5 : Calf Thymus Histone Preparation Nuclear Lysate with Human Histone H3 (tri methyl K4) peptide (**ab1342**) 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

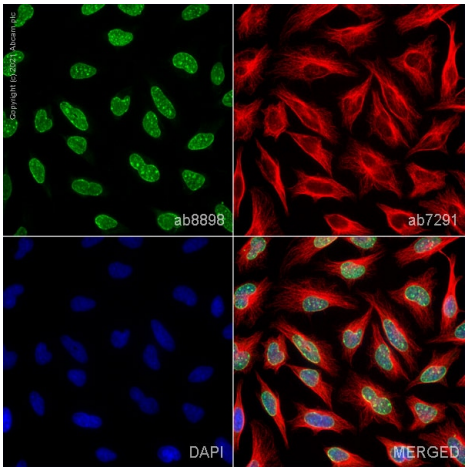
Lysates/proteins at 0.5 µg per lane.

Secondary
All lanes : IRDye 680 Conjugated Goat Anti-Rabbit IgG (H+L) at 1/10000 dilution

Performed under reducing conditions.

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

Lane 8 shows that Rabbit polyclonal to Histone H3 (tri methyl K9) is blocked by the addition of the immunizing peptide (**ab1773**). Cross-reactivity with Histone H3 peptide - tri methyl K27 (**ab1782**) is also shown in Lane 11.

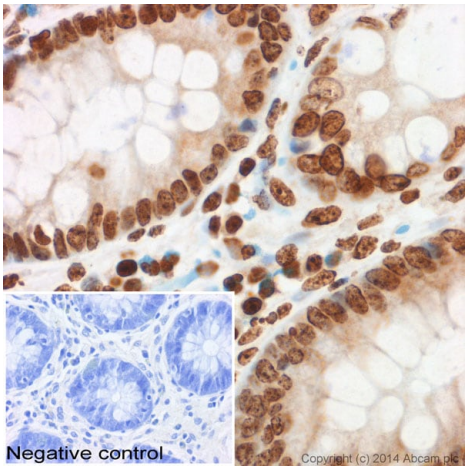


Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898)

ab8898 staining Histone H3 (tri methyl K9) in HeLa cells. The cells were fixed with 100% methanol (5 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 ab8898 at 0.5 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 4% paraformaldehyde (10 min).

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

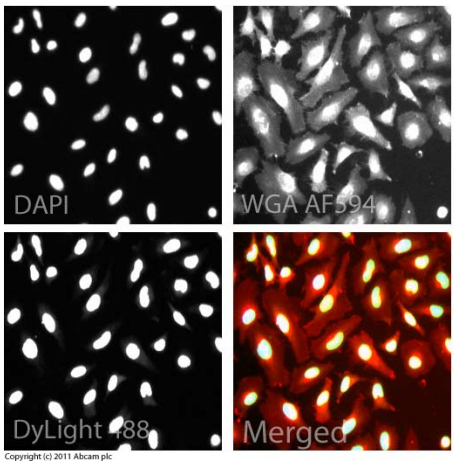


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898)

IHC image of ab8898 staining Histone H3 (tri methyl K9) in normal 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 ab8898, 1/400 dilution, 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.

*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (tri methyl K9) antibody - ChIP Grade (ab8898)

ICC/IF image of ab8898 stained HeLa cells. The cells were 100% methanol fixed (5 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 (ab8898, 0.1µg/ml) overnight at +4°C. The secondary antibody (green) was **ab96899**, a goat **anti-rabbit DyLight® 488** (IgG; H+L) used at a 1/250 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.

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