abcam

Product datasheet

Anti-Histone H3 (di methyl K79) antibody - ChIP Grade ab3594

15 Abreviews 221 References 画像数 9

製品の概要

製品名	Anti-Histone H3 (di methyl K79) antibody - ChIP Grade
製品の詳細	Rabbit polyclonal to Histone H3 (di methyl K79) - ChIP Grade
由来種	Rabbit
特異性	ab3594 detects a 17 kDa band in single lane Western Blot. Peptide inhibition in Western Blot hasn't been processed. Modification specificity is determined by Peptide Array. ab3594 binds strongly to the Histone H3 dii methyl K79. In Peptide Array ab3594 also partially binds to mono methyl K79 and tri methyl K79 peptides.
アプリケーション	**適用あり:** ChIP, WB, ICC/IF, IHC-P, PepArr
種交差性	**交差種:** Mouse, Cow, Human **交差が予測される動物種:** Pig, Saccharomyces cerevisiae, Caenorhabditis elegans, a wide range of other species, Mammals, Silk worm, Triticum aestivum
免疫原	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
ポジティブ・コントロール	ICC/IF: HeLa and HepG2 cells
特記事項	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ab3594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
ChIP	★★★★★ (2)	Use at an assay dependent concentration.
WB	★★★★★ (6)	Use at an assay dependent concentration. Predicted molecular weight: 15 kDa.
ICC/IF	★★★★★ (3)	Use a concentration of 0.1 - 1 µg/ml.
IHC-P	★★★★★ (1)	Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
PepArr		Use a concentration of 0.2 - 0.02 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.

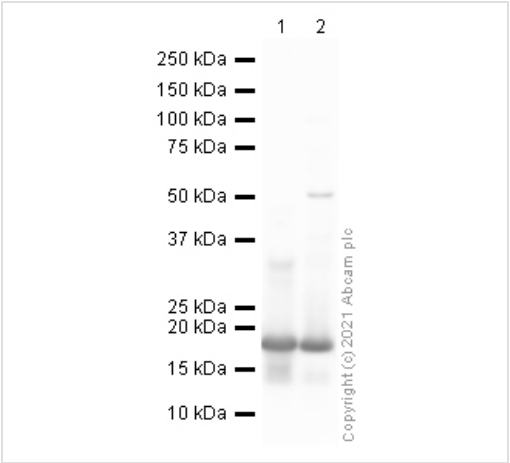
発生段階 Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

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



Western blot - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

All lanes : Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594) at 1 µg/ml

Lane 1 : CTH (Calf Thymus Histone) at 0.5 µg

Lane 2 : NIH 3T3 nuclear lysate (Triton enriched) at 10 µg

Secondary

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

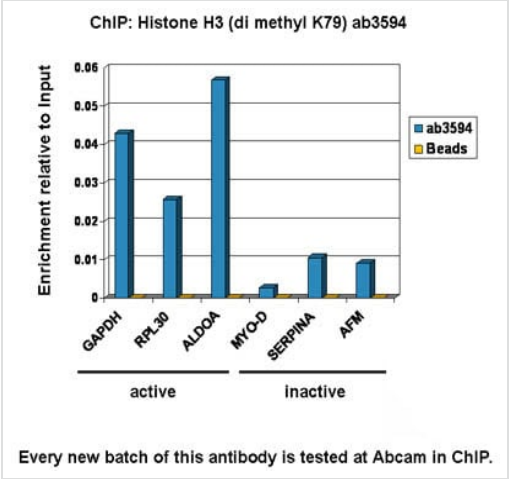
Predicted band size: 15 kDa

Observed band size: 17 kDa

Exposure time: 1 minute

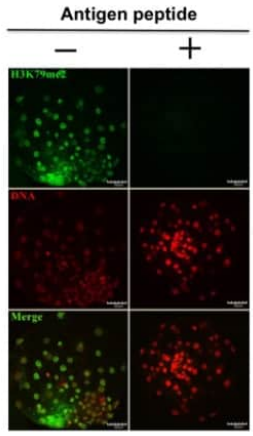
Gel type: MES

Blocking buffer: 2% BSA



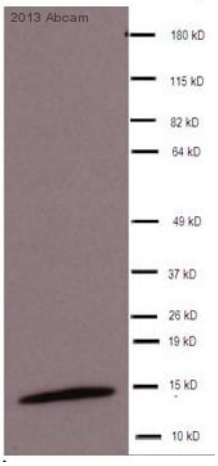
ChIP - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

Chromatin was prepared from U-2 OS (Human bone osteosarcoma epithelial cell line) 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 ab3594 (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). Primers and probes are located in the first kb of the transcribed region.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

Verification of H3K79me2 antibody specificity.
The commercial H3K79me2 primary antibody was preincubated with (+) or without (−) antigen peptide (Abcam, catalog no. **ab4556**, v/v = 5:1) at room temperature for 1.5 h before the incubation with IVF porcine blastocysts. H3K79me2 signals were observed in blastocysts using unabsorbed primary antibody. By contrast, H3K79me2 signals were absent in blastocysts using pre-absorbed primary antibody. H3K79me2 antibody was localized with Alexa Flour 488-conjugated secondary antibody (green). DNA was stained with propidium iodide (red). Bottom panels showed the merged images (yellow) between H3K79me2 signals (green) and DNA staining (red). Scale bars = 50μm.



Western blot - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

This image is courtesy of an anonymous Abreview

Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594) at 20 µg + HeLa cell lysate at 20 µg

Secondary

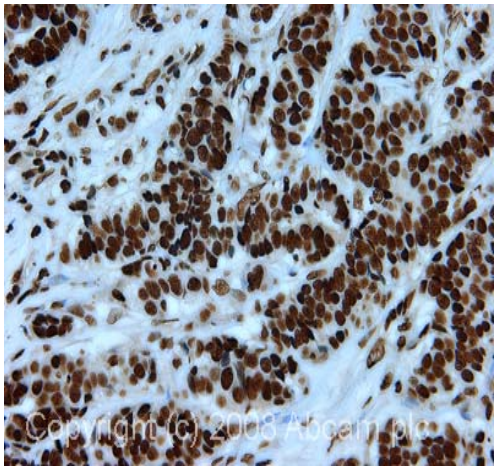
HRP-conjugated Goat anti-rabbit IgG polyclonal at 1/2000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

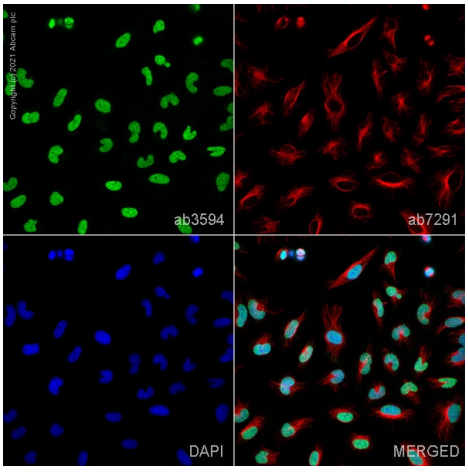
Predicted band size: 15 kDa
Observed band size: 15 kDa

Exposure time: 1 minute



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

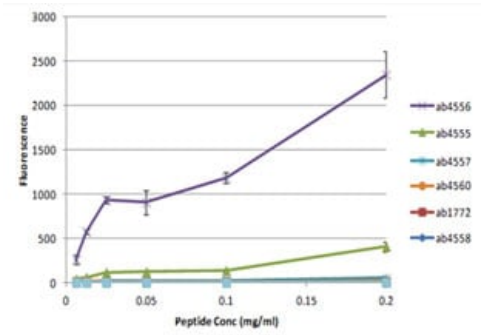
IHC image of Histone H3 (di methyl K79) staining in human breast carcinoma FFPE section, performed on a Bond™ system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab3594, 1µg/ml, for 8 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.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

ab3594 staining Histone H3 (Di Methyl K79) 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 ab3594 at 0.1µg/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).

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



Peptide Array - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

All batches of ab3594 are tested in Peptide Array against peptides to different Histone H3 modifications. Six dilutions of each peptide are printed on to the Peptide Array in triplicate and results are averaged before being plotted on to a graph. Results show strong binding to Histone H3 - di methyl K79 peptide (**ab4556**), indicating that this antibody specifically recognises the Histone H3 - di methyl K79 modification.

ab4556 - Histone H3 - di methyl K79

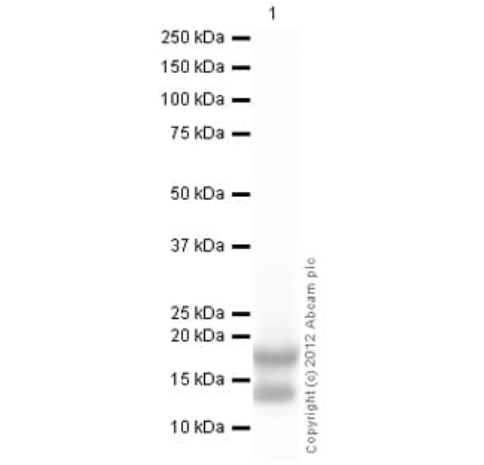
ab4555 - Histone H3 - mono methyl K79

ab4557 - Histone H3 - tri methyl K79

ab4560 - Histone H4 - di methyl K79

ab1772 - Histone H3 - di methyl K9

ab4558 - Histone H3 - unmodified



Western blot - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594) at 1 µg/ml + Calf Thymus Histone Preparation Nuclear Lysate (**ab121**) at 0.5 µg

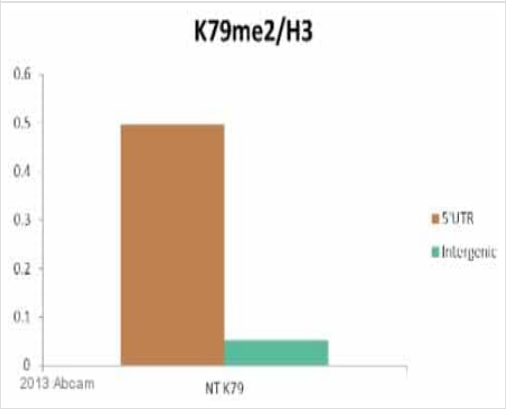
Secondary
Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/10000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

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

Exposure time: 30 seconds



ChIP - Anti-Histone H3 (di methyl K79) antibody - ChIP Grade (ab3594)

This image is courtesy of an anonymous Abreview

ChIP analysis using ab3594 binding Histone H3 in HeLa (Human epithelial adenocarcinoma cell line) cell nuclear lysate. Cells were cross-linked for 10 minutes with formaldehyde. Samples were incubated with primary antibody (5µg/µg chromatin) for 12 hours at 4°C. Protein binding was detected using real-time PCR.
Positive control: 5'UTR of transcribed gene.
Negative Control: Intergenic region.

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