

Product datasheet

Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade ab177870

Recombinant RabMAb

[8 Images](#)

Overview

Product name	Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade
Description	Rabbit monoclonal [EPR16595] to Histone H3 (acetyl K18) - ChIP Grade
Host species	Rabbit
Tested applications	Suitable for: WB, IHC-P, ICC/IF, ChIP, Dot blot
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	within Human Histone H3 aa 1-100 (acetyl K18). The exact sequence is proprietary. Database link: P68431
Positive control	WB: Whole cell lysates from HeLa and NIH/3T3 cells treated with 500 ng/ml Trichostatin A for 4 hours. IHC-P: Human colon, Mouse colon and Rat cerebral cortex tissues. ICC/IF: HeLa cells. ChIP: Chromatin prepared from HeLa cells.
General notes	Our RabMAb [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents .

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR16595
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab177870** in the following tested applications.

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

Application	Abreviews	Notes
WB		1/1000. Detects a band of approximately 15 kDa (predicted molecular weight: 15 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF		1/2000.
ChIP		Use at an assay dependent concentration. Use 2 µg.
Dot blot		1/1000.

Target

Function 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.

Sequence similarities Belongs to the histone H3 family.

Developmental stage Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

Post-translational modifications 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. 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

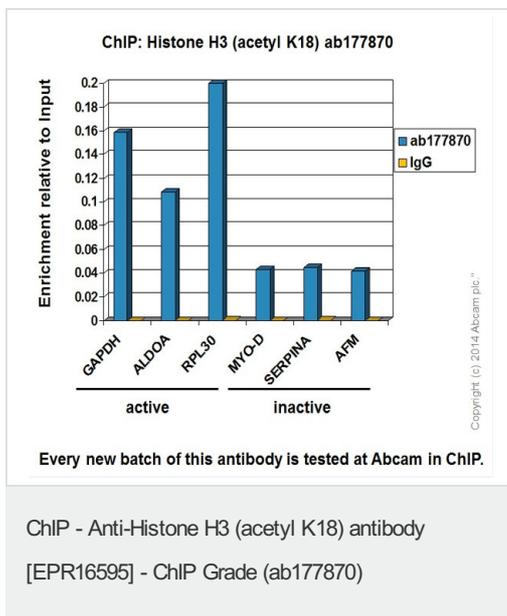
at Ser-11 (H3S10ph) by RPS6KA4 and RPS6KA5 is important during interphase because it enables the transcription of genes following external stimulation, like mitogens, stress, growth factors or UV irradiation and result in the activation of genes, such as c-fos and c-jun. Phosphorylation at Ser-11 (H3S10ph),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

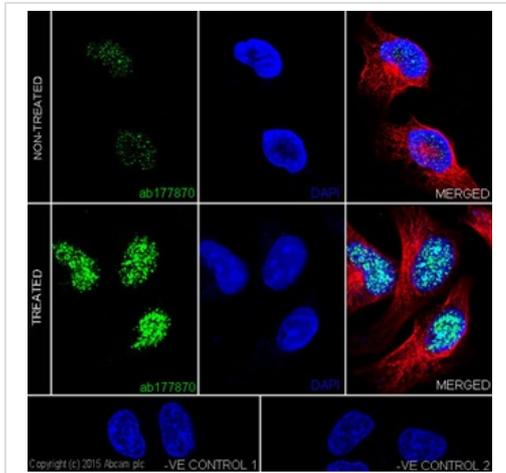
Cellular localization

Nucleus. Chromosome.

Images



Chromatin was prepared from HeLa (Human epithelial cells from cervix adenocarcinoma) 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 ab177870 (blue), and 20µl of Anti rabbit IgG sepharose beads. 2µg of Rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (SYBR approach). Primers and probes are located in the first kb of the transcribed region.



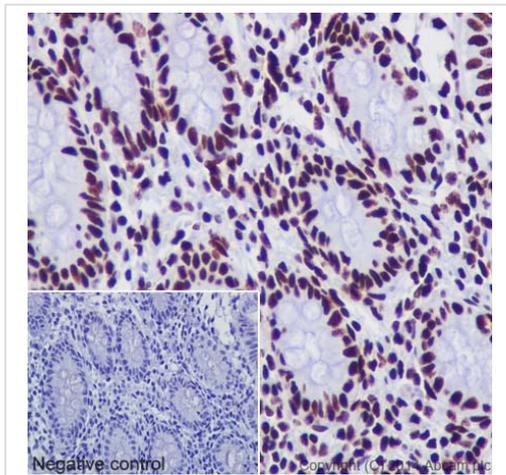
Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling Histone H3 (acetyl K18) with ab177870 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/400 dilution (green). Confocal image showing nuclear staining on HeLa cell line. Acetylation levels increased after treatment with Trichostatin A (500 ng/ml) for 4 hours. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (Anti-alpha Tubulin antibody [DM1A] - Loading Control) at 1/500 dilution and Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) preadsorbed (ab150120) at 1/500 dilution (red).

The negative controls are as follows;

-ve control 1: ab177870 at 1/2000 dilution followed by ab150120 at 1/500 dilution.

-ve control 2: ab7291 at 1/500 dilution followed by ab150077 at 1/400 dilution.

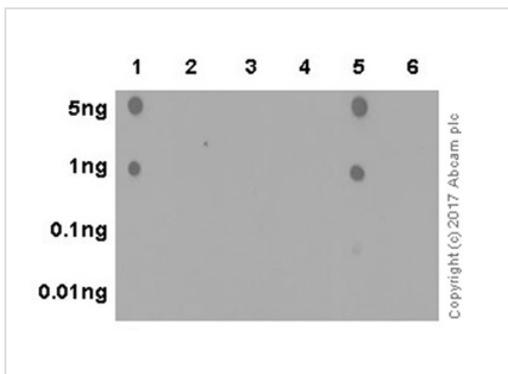


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Histone H3 (acetyl K18) with ab177870 at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nucleus staining on glandular epithelium of colon tissue is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Dot Blot - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

Dot blot analysis using ab177870 at 1/1000 dilution and Goat Anti-Rabbit IgG H&L (HRP) (ab97051) as secondary antibody at 1/100,000 dilution.

Lane 1: Histone H3(acetyl K4 + K9 + K14 + K18) peptide

Lane 2: Histone H3(acetyl K4) peptide

Lane 3: Histone H3(acetyl K9) peptide

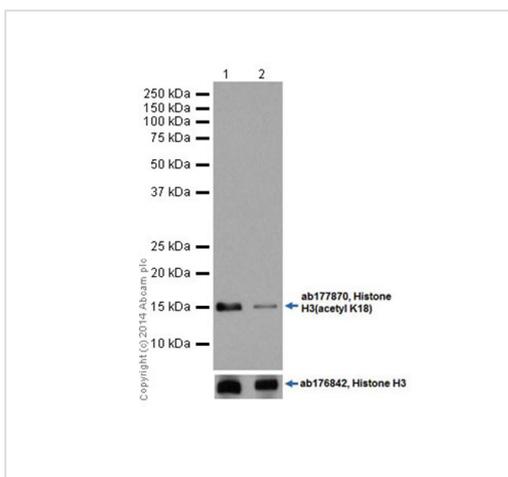
Lane 4: Histone H3(acetyl K14) peptide

Lane 5: Histone H3(acetyl K18) peptide

Lane 6: Histone H3 unmodified peptide

Blocking and Diluting buffer and concentration: 5% NFDm/TBST

Exposure time: 10 seconds



Western blot - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

All lanes : Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870) at 1/1000 dilution

Lane 1 : Whole cell lysates from HeLa (Human epithelial cells from cervix adenocarcinoma) cells treated with 500 ng/ml Trichostatin A for 4 hours

Lane 2 : Untreated HeLa whole cell lysates

Lysates/proteins at 10 µg per lane.

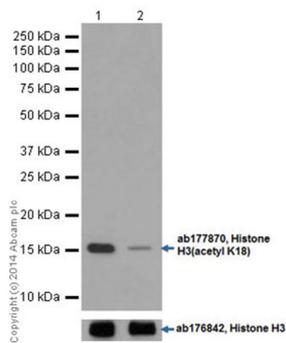
Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 15 kDa

Observed band size: 15 kDa

Blocking/Dilution buffer: 5% NFDm/TBST.



Western blot - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

All lanes : Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870) at 1/1000 dilution

Lane 1 : Whole cell lysate from NIH/3T3 (Mouse embryo fibroblast cells) treated with 500 ng/ml Trichostatin A for 4 hours

Lane 2 : Whole cell lysates from untreated NIH/3T3

Lysates/proteins at 10 µg per lane.

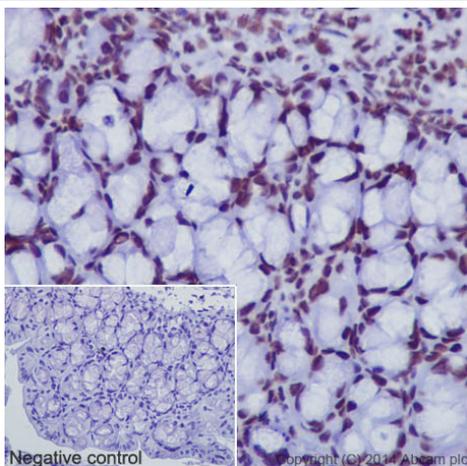
Secondary

All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 15 kDa

Observed band size: 15 kDa

Blocking/Dilution buffer: 5% NFDM/TBST.

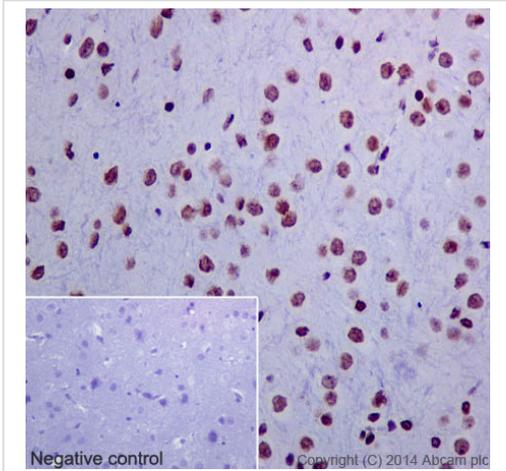


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labeling Histone H3 (acetyl K18) with ab177870 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nuclear staining on glandular epithelium of mouse colon tissue is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemical analysis of paraffin-embedded Rat cerebral cortex tissue labeling Histone H3 (acetyl K18) with ab177870 at 1/500 dilution, followed by prediluted Goat Anti-Rabbit IgG H&L (HRP). Nuclear staining on Rat cerebral cortex tissue is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody, secondary antibody is prediluted Goat Anti-Rabbit IgG H&L (HRP).

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EPR16595] - ChIP Grade (ab177870)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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.com/abpromise> or contact our technical team.

Terms and conditions

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