abcam

Product datasheet

Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade ab40888

Recombinant RabMAb

1 Abreviews 14 References 13 Images

Overview

Product name	Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade
Description	Rabbit monoclonal [EP959Y] to Histone H3 (acetyl K18) - ChIP Grade
Host species	Rabbit
Specificity	This antibody only detects Histone H3 when acetylated on Lysine 18.
Tested applications	**Suitable for:** ICC/IF, ChIP, ChIP-sequencing, WB, IHC-P
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Histone H3 aa 1-100 (acetyl K18). The exact sequence is proprietary.
Positive control	WB: C6, NIH/3T3 and HeLa cells treated with 500ng/ml Trichostatin A for 4 hours. IHC-P: Human breast carcinoma and endometrium cancer tissue. Mouse liver and rat stomach tissue. ICC/IF: HeLa cells. ChIP: Chromatin prepared from HeLa cells. ChIP-seq: Chromatin prepared from HeLa cells.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information **see here**. 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. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

Purity	Protein A purified
Clonality	Monoclonal
Clone number	EP959Y
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab40888 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
ICC/IF	★★★★★ (1)	1/1000. **For unpurified use at 1/100-1/250**
ChIP		Use 5 µg for 25 µg of chromatin.
ChIP-sequencing		Use 4µg for 10^7 cells.
WB		1/20000. Detects a band of approximately 17 kDa (predicted molecular weight: 17 kDa). **For unpurified use at 1/500**
IHC-P		1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

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.

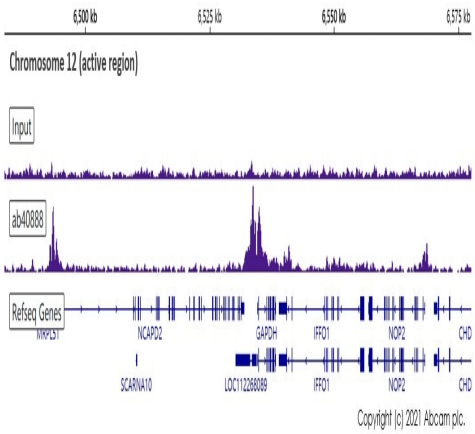
Developmental stage Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

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 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.

Cellular localization Nucleus. Chromosome.

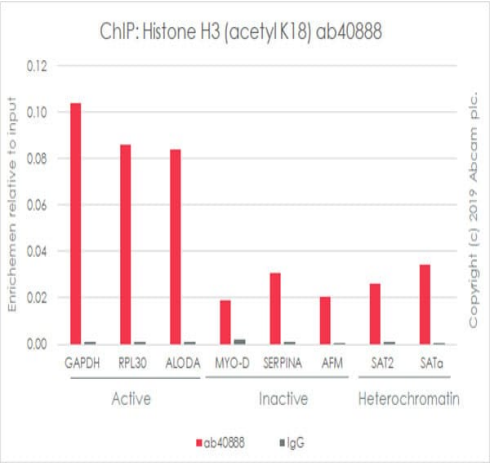
Images



ChIP-sequencing - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

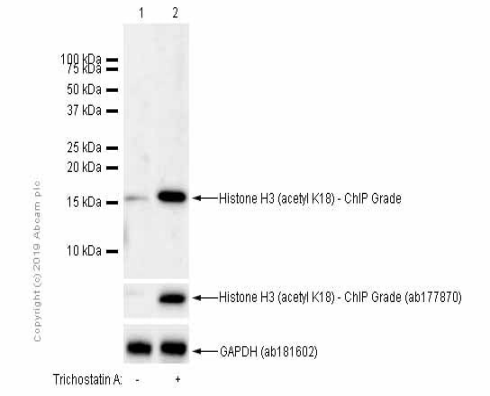
Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 4 µg of ab40888 [EP959Y]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded **here**.



ChIP - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol*. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25 µg of chromatin, 5 µg of ab40888 (red), and 20 µl of Protein A/G sepharose beads. 5 µg of rabbit normal IgG was added to the beads control (gray). 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 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

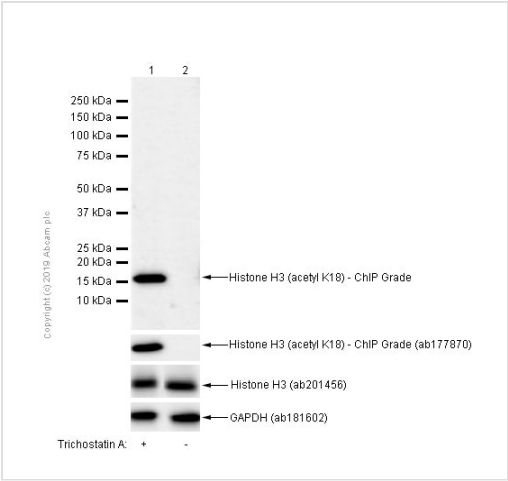
All lanes : Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888) at 1/20000 dilution (Purified)

Lane 1 : HeLa (Human cervix adenocarcinoma epithelial cell) whole cell lysates
Lane 2 : HeLa (Human cervix adenocarcinoma epithelial cell) treated with 500ng/ml Trichostatin A for 4 hours whole cell lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

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



Western blot - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

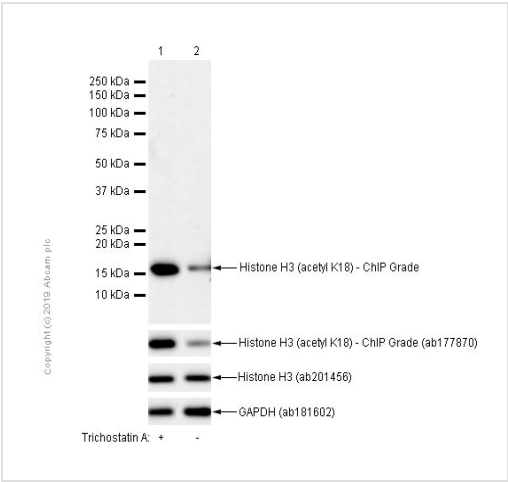
All lanes : Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888) at 1/50000 dilution (Purified)

Lane 1 : NIH/3T3 (Mouse embryonic fibroblast) treated with 500ng/ml Trichostatin A for 4 hours whole cell lysates
Lane 2 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

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



Western blot - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

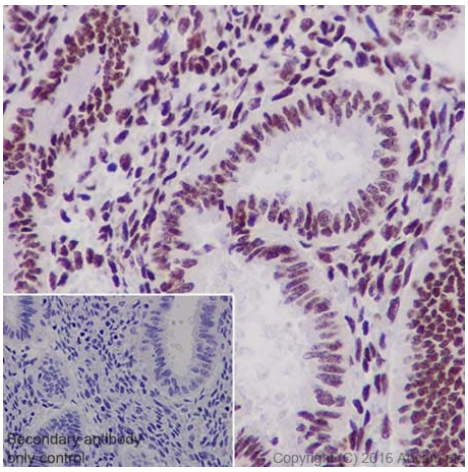
All lanes : Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888) at 1/50000 dilution (Purified)

Lane 1 : C6 (Rat glial tumor glial cell) treated with 500ng/ml Trichostatin A for 4 hours whole cell lysates
Lane 2 : C6 (Rat glial tumor glial cell) whole cell lysates

Lysates/proteins at 15 µg per lane.

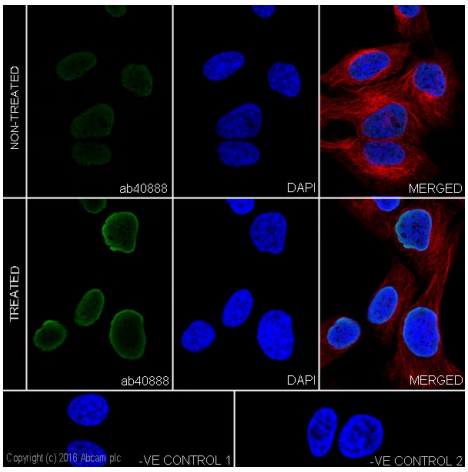
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

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



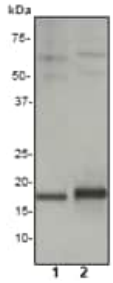
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human endometrium cancer tissue sections labeling Histone H3 with purified ab40888 at 1/4000 dilution (0.23 µg/ml). Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human cervix adenocarcinoma epithelial cell) treated with 500ng/ml Trichostatin A for 4 hours cells labeling Histone H3 with purified ab40888 at 1/1000 dilution (0.9 µg/ml). Cells were fixed in 4% Paraformaldehyde and permeabilized with 0.1% tritonX-100. Cells were counterstained with **ab195889** Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) at 1/200 (2.5 µg/ml) dilution. Goat anti rabbit IgG (Alexa Fluor® 488, **ab150077**) was used as the secondary antibody at 1/1000 (2 µg/ml) dilution. DAPI (blue) was used as nuclear counterstain. PBS instead of the primary antibody was used as the secondary antibody only control.



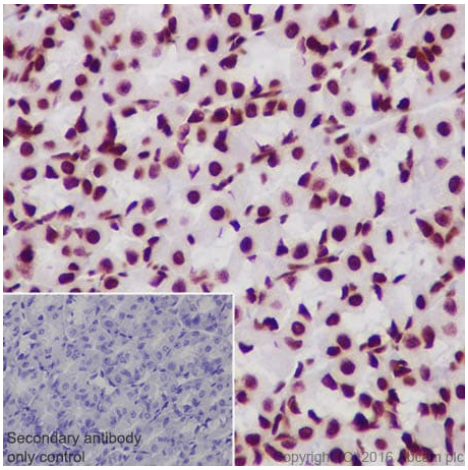
Western blot - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

All lanes : Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888) at 1/50000 dilution

Lane 1 : C6 cell lysate, untreated.
Lane 2 : C6 cell lysate, treated with TSA.

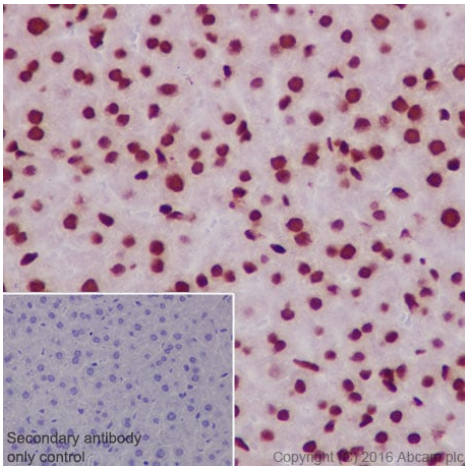
Lysates/proteins at 10 µg per lane.

Predicted band size: 17 kDa
Observed band size: 17 kDa
Additional bands at: 50 kDa (possible non-specific binding), 60 kDa (possible non-specific binding)



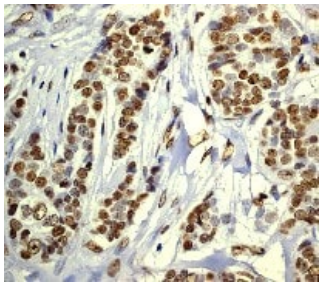
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of rat stomach tissue sections labeling Histone H3 with purified ab40888 at 1/4000 dilution (0.23 µg/ml). Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



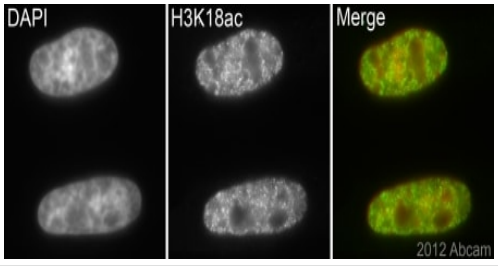
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse liver tissue sections labeling Histone H3 with purified ab40888 at 1/4000 dilution (0.23 µg/ml). Perform heat mediated antigen retrieval using **ab93684** (Tris/EDTA buffer, pH 9.0). ImmunoHistoProbe one step HRP Polymer (ready to use) was used as the secondary antibody. Negative control: PBS instead of the primary antibody. Hematoxylin was used as a counterstain.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

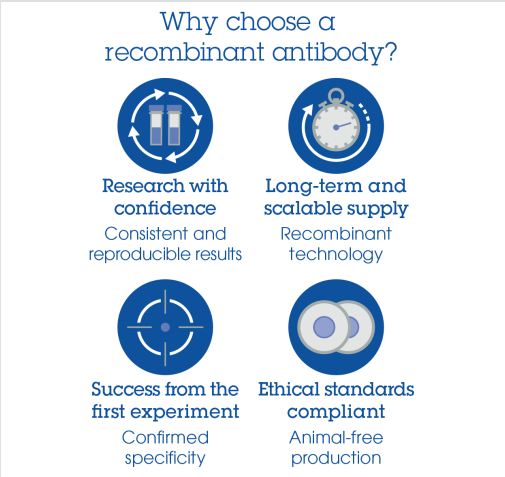
ab40888 diluted 1:100, staining acetylated histone H3 on human breast carcinoma sections.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

Image courtesy of an Abreview submitted by Dr. Kirk McManus, Univ. of Manitoba/Cancer Care MICB, Canada

ab40888 (1/500) staining Histone H3 (acetyl K18) in asynchronous HeLa cells (green). Cells were fixed in paraformaldehyde, permeabilised with 0.5% Triton X-100/PBS and counterstained with DAPI in order to highlight the nucleus (red). For further experimental details please refer to Abreview.



Anti-Histone H3 (acetyl K18) antibody [EP959Y] - ChIP Grade (ab40888)

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