abcam

Product datasheet

Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade ab9048

14 Abreviews 106 References 4 Images

Overview

Product name	Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade
Description	Rabbit polyclonal to Histone H3 (mono methyl K36) - ChIP Grade
Host species	Rabbit
Specificity	Specific for human Histone H3 mono methyl K36. Shows partial cross-reactivity with di-methyl K36 (please see Western Blot image). This antibody may not be suitable for experiments on yeast lysate. Although the antibody is specifically blocked using the immunising peptide, customer feedback indicates that it detects a band using S. cerevisiae K36 point mutants. We welcome further customer feedback.
Tested applications	**Suitable for:** WB, ChIP, ICC/IF
Species reactivity	**Reacts with:** Human **Predicted to work with:** Mouse, Cow, Pig, Saccharomyces cerevisiae, Arabidopsis thaliana, Drosophila melanogaster, Mammals
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	ICC/IF: HeLa cells
General notes	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

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 or -80°C. Avoid freeze / thaw cycle.
Storage buffer	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.

Purity Immunogen affinity purified

Clonality Polyclonal

Isotype IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab9048 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	★★☆☆☆ (4)	1/1000.
ChIP	★★★★☆ (3)	Use 2 µg for 25 µg of chromatin.
ICC/IF	★★★★★ (1)	Use a concentration of 0.1 µg/ml.

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.

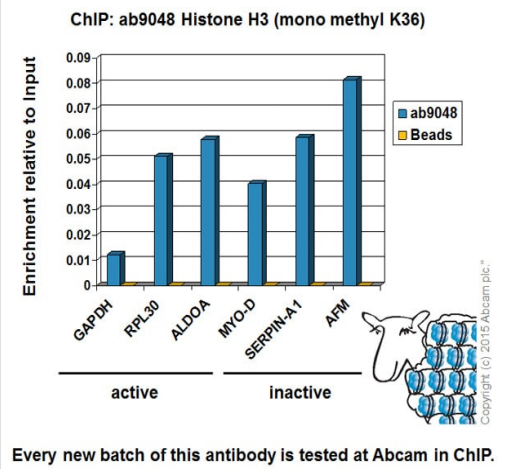
Developmental stage Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

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.

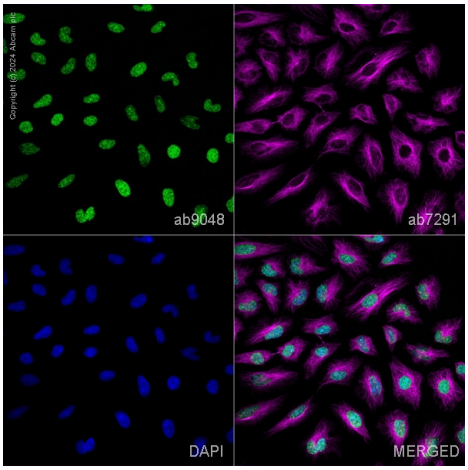
Cellular localization Nucleus. Chromosome.

Images



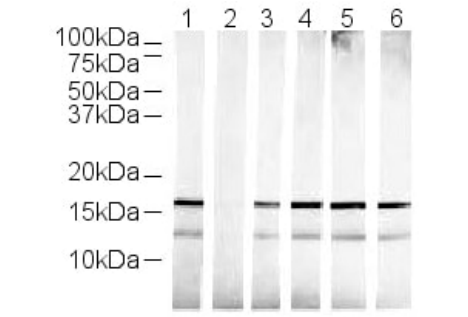
ChIP - Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade (ab9048)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 2µg of ab9048 (blue), and 20µl 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 (mono methyl K36) antibody - ChIP Grade (ab9048)

ab9048 staining Histone H3 (mono methyl K36) 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 ab9048 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 magenta). 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.



Western blot - Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade (ab9048)

All lanes : Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade (ab9048) at 1/500 dilution

Lane 1 : Histone prep
Lane 2 : Histone prep with Human Histone H3 (mono methyl K36) peptide (**ab1783**) at 1 µg/ml
Lane 3 : Histone prep with Human Histone H3 (di methyl K36) peptide (**ab1784**) at 1 µg/ml
Lane 4 : Histone prep with Human Histone H3 (tri methyl K36) peptide (**ab1785**) at 1 µg/ml
Lane 5 : Histone prep with Human Histone H3 (unmodified) peptide (**ab2623**) at 1 µg/ml
Lane 6 : Histone prep with Human Histone H3 (mono methyl K4) peptide (**ab1340**) at 1 µg/ml

Lysates/proteins at 0.5 µg per lane.

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

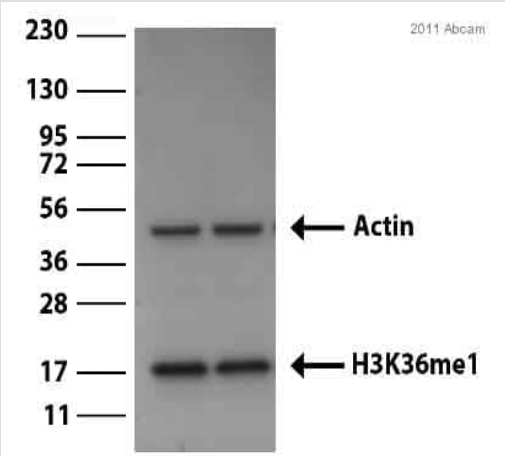
Secondary ab: Alexa Fluor 680 Goat anti-rabbit IgG

0.5µg histone prep used per lane

Primary antibody:

Lane 1: ab9048 (Histone H3 Mono Methyl K36) 1/500
Lane 2: ab9048 (Histone H3 Mono Methyl K36) 1/500 + **ab1783** (ab9048) (Histone H3 Mono Methyl K36) peptide 1µg/ml
Lane 3: ab9048 (Histone H3 Mono Methyl K36) 1/500 + **ab1794** (**ab9049**) (Histone H3 Di Methyl K36) peptide 1µg/ml
Lane 4: ab9048 (Histone H3 Mono Methyl K36) 1/500 + **ab1785** (**ab9050**) (Histone H3 Tri Methyl K36) peptide 1µg/ml
Lane 5: ab9048 (Histone H3 Mono Methyl K36) 1/500 + **ab2623** (Histone H3 (23-34) – unmodified) peptide 1µg/ml
Lane 6: ab9048 (Histone H3 Mono Methyl K36) 1/500 + **ab1340** (**ab8895**) (Histone H3 Mono methyl K4) peptide 1µg/ml

ab9048 specifically recognise



Western blot - Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade (ab9048)
Image courtesy of an anonymous Abreview.

All lanes : Anti-Histone H3 (mono methyl K36) antibody - ChIP Grade (ab9048) at 1/1400 dilution

All lanes : Whole cell lysate prepared from Drosophila BG3 cells

Lysates/proteins at 500000 cells per lane.

Secondary
All lanes : HRP donkey anti-rabbit monoclonal at 1/20000 dilution

Developed using the ECL technique.

Observed band size: 17,42 kDa

Exposure time: 30 seconds

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