abcam

Product datasheet

Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody ab231674

5 Images

Overview

Product name	Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody
Description	Rabbit polyclonal to Histone H3 (asymmetric di methyl R17, acetyl K18)
Host species	Rabbit
Tested applications	**Suitable for:** WB, ChIP, Dot blot, ICC/IF, CHIPseq
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Synthetic peptide corresponding to Human Histone H3 (asymmetric di methyl R17, acetyl K18) conjugated to keyhole limpet haemocyanin. Database link: P68431
Positive control	ICC/IF: NIH/3T3 cells. WB: Whole cell and histone extracts from HeLa cells. ChIP: HeLa cells.

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	Preservatives: 0.05% Sodium azide, 0.05% Proclin Constituent: PBS
Purity	Affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab231674** 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.

Application	Abreviews	Notes
ChIP		Use 1-10µg for 10^6 cells.
Dot blot		1/20000.
ICC/IF		1/500.
CHIPseq		Use at an assay dependent concentration.

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.

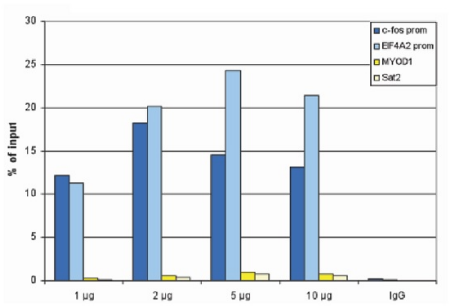
Post-translational modifications

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.

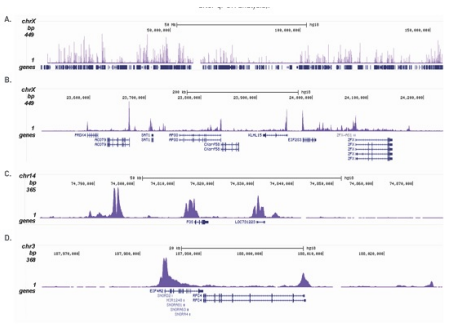
Cellular localization Nucleus. Chromosome.

Images



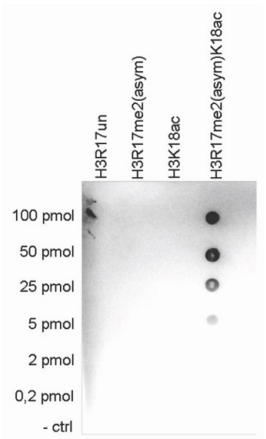
ChIP - Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674)

ChIP assays were performed using HeLa (Human epithelial cell line from cervix adenocarcinoma) cells, ab231674 and optimized PCR primer pairs for qPCR. ChIP was performed using sheared chromatin from 1,000,000 cells. A titration consisting of 1, 2, 5 and 10 µg of antibody per ChIP experiment was analyzed. IgG (2 µg/IP) was used as a negative IP control. Quantitative PCR was performed with primers for the promoters of the active EIF4A2 and c-fos genes, used as positive controls and for the inactive MYOD1 gene and the Sat2 satellite repeat, used as negative controls. Image shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).



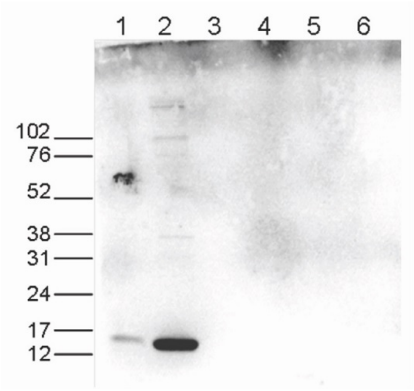
CHIPseq - Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674)

ChIP was performed as described above using 1 µg of ab231674. The IP'd DNA was subsequently analysed on an Illumina Genome Analyzer. Library preparation, cluster generation and sequencing were performed according to the manufacturer's instructions. The 36 bp tags were aligned to the human genome using the ELAND algorithm. Image shows the peak distribution along the complete human X-chromosome and a zoomin to a 500 kb region (2A and B), and in two regions on chromosome 14 and 3 surrounding the c-fos and EIF4A2 positive control genes (2C and D, respectively).



Dot Blot - Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674)

Dot Blot analysis was performed with peptides containing other histone modifcations and the unmodifed H3R17K18. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. ab231674 was used at a dilution of 1:20,000.



Western blot - Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674)

All lanes : Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell extracts at 25 µg
Lane 2 : HeLa (Human epithelial cell line from cervix adenocarcinoma) histone extracts at 15 µg
Lane 3 : Recombinant histone H2A at 1 µg
Lane 4 : Recombinant histone H2B at 1 µg
Lane 5 : Recombinant histone H3 at 1 µg
Lane 6 : Recombinant histone H4 at 1 µg

Dilution buffer: TBS-Tween containing 5% skimmed milk.

Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (asymmetric di methyl R17, acetyl K18) antibody (ab231674)

NIH/3T3 (Mouse embryo fibroblast cell line) cells stained for Histone H3 (asymmetric di methyl R17, acetyl K18) using ab231674 at a dilution of 1/500 in ICC/IF.

Cells were fxed with 4% formaldehyde for 10 minutes and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA. Secondary used is an Alexa Fluor®488-conjugated Anti-Rabbit. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.

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