# abcam

# Product datasheet

# Anti-Histone H2B (acetyl K12) antibody - ChIP Grade ab195494

2 References 5 Images

Overview

Product name Anti-Histone H2B (acetyl K12) antibody - ChIP Grade

**Description** Rabbit polyclonal to Histone H2B (acetyl K12) - ChIP Grade

Host species Rabbit

Specificity Due to sequence homology we expect this antibody to detect K12 acetylation on multiple H2B

variants

Tested applications Suitable for: ICC/IF, Dot blot, ChIP, WB, ChIP-sequencing

Species reactivity Reacts with: Human, Recombinant fragment

Immunogen Synthetic peptide corresponding to Human Histone H2B (acetyl K12) conjugated to keyhole

limpet haemocyanin. Peptide surrounding K12.

Database link: Q5QNW6

Positive control Human HeLa cells and HeLaS3 cells. HeLa whole cell and histone extracts.

**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 long

term. Avoid freeze / thaw cycle.

**Storage buffer** Preservatives: 0.05% Sodium azide, 0.05% Proclin 300

Constituent: 99% PBS

Purity Protein A purified

**Clonality** Polyclonal

**Isotype** IgG

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#### **Applications**

### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab195494 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		1/500.
Dot blot		1/5000.
ChIP		Use at an assay dependent concentration. 0.5 - 5 μg per ChIP
WB		1/1000. Predicted molecular weight: 14 kDa.
ChIP-sequencing		Use at an assay dependent concentration. 0.5 - 5 μg per ChIP

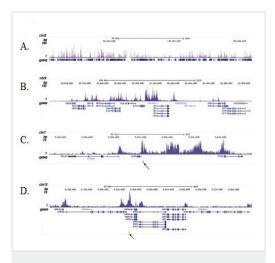
# **Target**

#### Relevance

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. Subunit structure The nucleosome is a histone octamer containing two molecules each of H2A, H2B, H3 and H4 assembled in one H3-H4 heterotetramer and two H2A-H2B heterodimers. The octamer wraps approximately 147 bp of DNA. Post-translational modification Monoubiquitination at Lys-35 (H2BK34Ub) by the MSL1/MSL2 dimer is required for histone H3 'Lys-4' (H3K4me) and 'Lys-79' (H3K79me) methylation and transcription activation at specific gene loci, such as HOXA9 and MEIS1 loci. Similarly, monoubiquitination at Lys-121 (H2BK120Ub) by the RNF20/40 complex gives a specific tag for epigenetic transcriptional activation and is also prerequisite for histone H3 'Lys-4' and 'Lys-79' methylation. It also functions cooperatively with the FACT dimer to stimulate elongation by RNA polymerase II. H2BK120Ub also acts as a regulator of mRNA splicing: deubiquitination by USP49 is required for efficient cotranscriptional splicing of a large set of exons. Phosphorylation at Ser-37 (H2BS36ph) by AMPK in response to stress promotes transcription. Phosphorylated on Ser-15 (H2BS14ph) by STK4/MST1 during apoptosis; which facilitates apoptotic chromatin condensation. Also phosphorylated on Ser-15 in response to DNA double strand breaks (DSBs), and in correlation with somatic hypermutation and immunoglobulin class-switch recombination. GlcNAcylation at Ser-113 promotes monoubiquitination of Lys-121. It fluctuates in response to extracellular glucose, and associates with transcribed genes. Crotonylation (Kcr) is specifically present in male germ cells and marks testis-specific genes in post-meiotic cells, including X-linked genes that escape sex chromosome inactivation in haploid cells. Crotonylation marks active promoters and enhancers and confers resistance to transcriptional repressors. It is also associated with post-meiotically activated genes on autosomes.

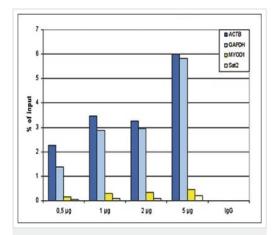
#### **Cellular localization**

Nuclear



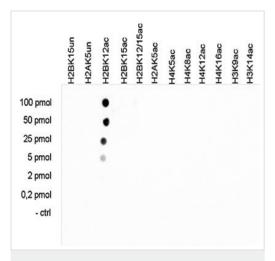
ChIP-sequencing - Anti-Histone H2B (acetyl K12) antibody - ChIP Grade (ab195494)

ChIP-seq analysis using Human HeLaS3 cells, labeling Histone H2B type 1-C/E/F/G/I (acetyl K12) with ab195494 at 0.5 µg and optimized PCR primer sets for qPCR as described above. The 51 bp tags were aligned to the Human genome using the BWA algorithm. The figure shows the enrichment along the complete sequence and a 1 Mb region of the X-chromosome (A and B) and in genomic regions of chromosome 7, surrounding the ACTB gene, and of chromosome 12, surrounding the GAPDH gene (C and D). The position of the amplicon used for ChIP-qPCR is indicated by an arrow.



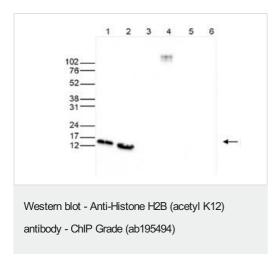
ChIP - Anti-Histone H2B (acetyl K12) antibody - ChIP Grade (ab195494)

ChIP analyis using Human HeLa cells, labeling Histone H2B type 1-C/E/F/G/I (acetyl K12) with ab195494 and optimized PCR primer sets for qPCR. ChIP was performed using sheared chromatin from 1.5 million cells. A titration of the antibody consisting of 0.5, 1, 2 and 5 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. QPCR was performed for a region approximately 1 kb upstream of the GAPDH and ACTB promoters, used as positive controls, and for the coding region of the inactive MYOD1 gene and the Sat2 satellite repeat, used as negative controls, respectively. The graph shows the recovery, expressed as a % of input (the relative amount of immunoprecipitated DNA compared to input DNA after qPCR analysis).



Dot Blot - Anti-Histone H2B (acetyl K12) antibody - ChIP Grade (ab195494)

Dot Blot analysis of peptides containing the unmodified H2B and other histone modifications, labeling Histone H2B type 1-C/E/F/G/I (acetyl K12) with ab195494 at 1/5000 dilution. 0.2-100 pmol of the peptide containing the respective modifications were spotted onto the membrane.



**All lanes :** Anti-Histone H2B (acetyl K12) antibody - ChIP Grade (ab195494) at 1/1000 dilution (in TBS-Tween containing 5% skimmed milk)

Lane 1 : HeLa whole cell extracts at 25  $\mu g$ 

Lane 2: HeLa histone extracts at 15 µg

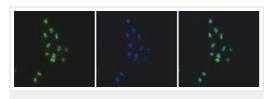
Lane 3: Recombinant H2A at 1 µg

Lane 4: Recombinant H2B at 1 µg

Lane 5: Recombinant H3 at 1 µg

Lane 6: Recombinant H4 at 1 µg

Predicted band size: 14 kDa



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2B (acetyl K12) antibody - ChIP Grade (ab195494) Immunofluorescent analysis of HeLa cells labeling Histone H2B type 1-C/E/F/G/I (acetyl K12) with ab195494 at 1/500 dilution in blocking solution followed by an anti-rabbit antibody conjugated to Alexa488 (left), DAPI (center) or merged (right). Cells were fixed with 4% formaldehyde for 10 minutes and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA.

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

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