

Product datasheet

Anti-Histone H4 (acetyl K5 + K8 + K12) antibody ab233193

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Overview

Product name	Anti-Histone H4 (acetyl K5 + K8 + K12) antibody
Description	Rabbit polyclonal to Histone H4 (acetyl K5 + K8 + K12)
Host species	Rabbit
Tested applications	Suitable for: ChIP-sequencing, WB, ICC/IF, Dot blot, ChIP
Species reactivity	Reacts with: Human, Recombinant fragment
Immunogen	Synthetic peptide corresponding to Human Histone H4 (acetyl K5 + K8 + K12) conjugated to keyhole limpet haemocyanin. Database link: P62805
Positive control	ChIP: Chromatin from K562 cells. ChIPseq: Chromatin from K562 cells. WB: HeLa whole cell and histone extracts. ICC/IF: HeLa cells.
General notes	<p>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.</p> <p>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</p>

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: PBS
Purity	Affinity purified
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab233193 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
ChIP-sequencing		Use at an assay dependent concentration. Use 0.5 µg.
WB		1/1000.
ICC/IF		1/500.
Dot blot		1/20000.
ChIP		Use at an assay dependent concentration. Use 0.5 - 1 µg per IP.

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 H4 family.

Post-translational modifications

Acetylation at Lys-6 (H4K5ac), Lys-9 (H4K8ac), Lys-13 (H4K12ac) and Lys-17 (H4K16ac) occurs in coding regions of the genome but not in heterochromatin.

Citrullination at Arg-4 (H4R3ci) by PAD4 impairs methylation.

Monomethylation and asymmetric dimethylation at Arg-4 (H4R3me1 and H4R3me2a, respectively) by PRMT1 favors acetylation at Lys-9 (H4K8ac) and Lys-13 (H4K12ac).

Demethylation is performed by JMJD6. Symmetric dimethylation on Arg-4 (H4R3me2s) by the PRDM1/PRMT5 complex may play a crucial role in the germ-cell lineage.

Monomethylated, dimethylated or trimethylated at Lys-21 (H4K20me1, H4K20me2, H4K20me3).

Monomethylation is performed by SET8. Trimethylation is performed by SUV420H1 and SUV420H2 and induces gene silencing.

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. Monoubiquitinated at Lys-92 of histone H4 (H4K91ub1) in response to DNA damage.

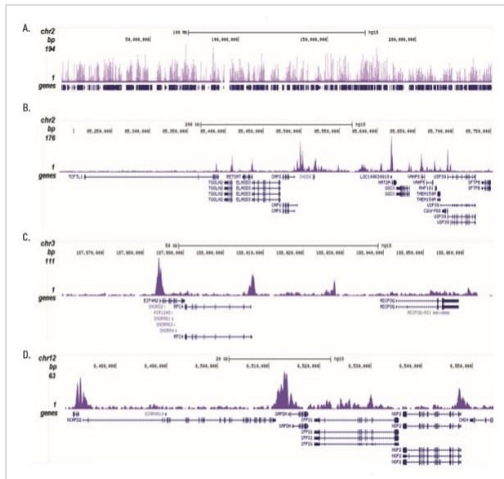
The exact role of H4K91ub1 in DNA damage response is still unclear but it may function as a licensing signal for additional histone H4 post-translational modifications such as H4 Lys-21 methylation (H4K20me).

Sumoylated, which is associated with transcriptional repression.

Cellular localization

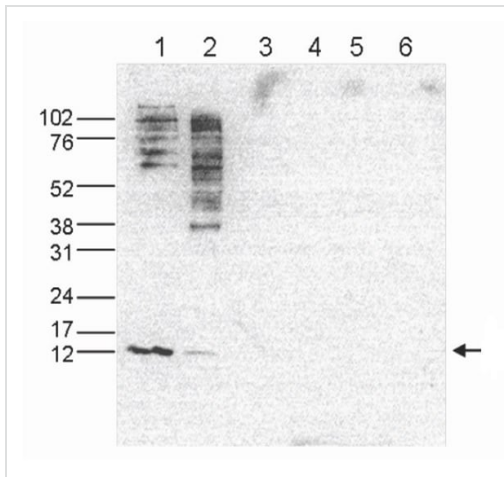
Nucleus. Chromosome.

Images



ChIP-sequencing - Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193)

ChIP was performed with 0.5 μ g ab233193 on sheared chromatin from 100,000 K562 cells. 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. The figure shows the signal distribution along the complete length of chromosome 2 (figure A) and a zoomin to a 600 kb region (figure B). Figure C and D show the enrichment in two genomic regions on chromosome 3 and 12, respectively, containing EIF4A2 and GAPDH positive controls.



Western blot - Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193)

All lanes : Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193) at 1/1000 dilution

Lane 1 : HeLa (human epithelial cell line from cervix

adenocarcinoma) whole cell extract at 25 μ g

Lane 2 : HeLa histone extract at 15 μ g

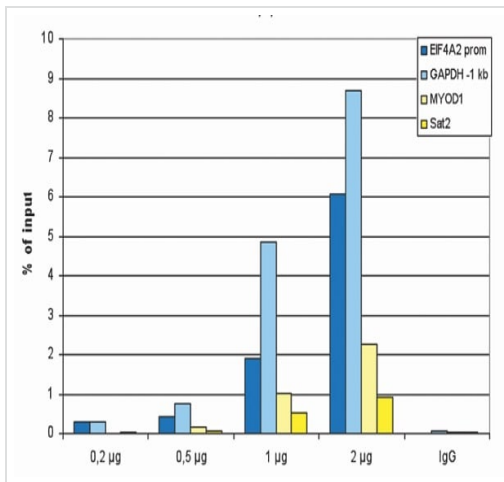
Lane 3 : Recombinant histone H2A at 1 μ g

Lane 4 : Recombinant histone H2B at 1 μ g

Lane 5 : Recombinant histone H3 at 1 μ g

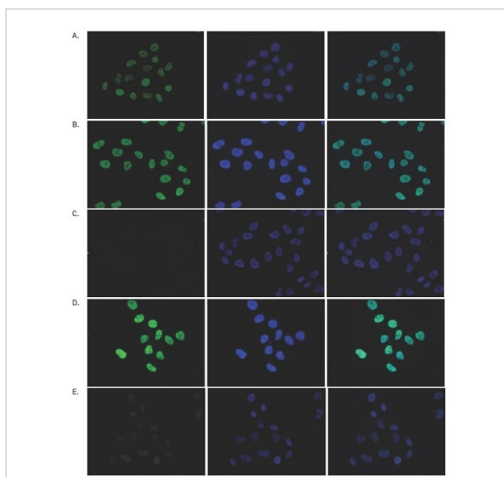
Lane 6 : Recombinant histone H4 at 1 μ g

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



ChIP - Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193)

ChIP assays were performed using human K562 (Human chronic myelogenous leukemia cell line from bone marrow) cells, ab233193 and optimized PCR primer sets for qPCR. ChIP was performed on sheared chromatin from 100,000 cells. A titration of the antibody consisting of 0.2, 0.5, 1 and 2 µg per ChIP experiment was analysed. IgG (1 µg/IP) was used as negative IP control. QPCR was performed with primers for promoter of the active gene EIF4A2 and for a region 1 kb upstream of the GAPDH gene, used as positive controls, and for the inactive MYOD1 gene and the Sat2 satellite repeat region 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).



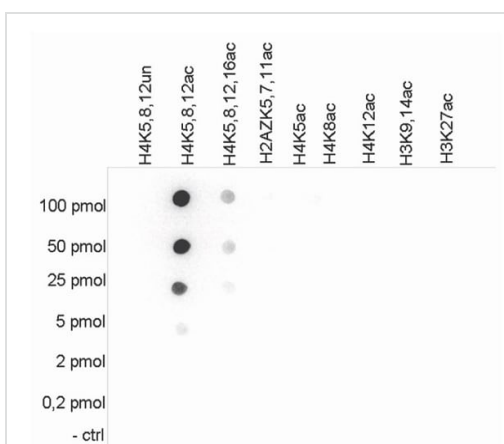
Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193)

HeLa (Human epithelial cell line from cervix adenocarcinoma) cells were stained with ab233193 and with DAPI.

Cells were fixed with 4% formaldehyde for 10 minutes and blocked with PBS/TX-100 containing 5% normal goat serum and 1% BSA.

Figure A: Cells were immunofluorescently labeled with ab233193 (left) diluted 1/500 in blocking solution followed by an anti-rabbit antibody conjugated to Alexa[®]488. The middle panel shows staining of the nuclei with DAPI. A merge of the two stainings is shown on the right.

Figure B, C, D and E: Staining of the cells with ab233193 after incubation of the antibody with 10 ng/µl of the following blocking peptides: H4K5,8,12 unmodified (B), H4K5,8,12ac (C), H2A.ZK5,7,11ac (D) and H4K5,8,12,16ac (E).



Dot Blot - Anti-Histone H4 (acetyl K5 + K8 + K12) antibody (ab233193)

To test the cross reactivity of ab233193 a Dot Blot analysis was performed with peptides containing other histone modifications and the unmodified H4. One hundred to 0.2 pmol of the respective peptides were spotted on a membrane. ab233193 was used at a dilution of 1:20,000.

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