abcam

Product datasheet

Anti-Histone H2A antibody - ChIP Grade ab13923

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Overview

Product name	Anti-Histone H2A antibody - ChIP Grade
Description	Rabbit polyclonal to Histone H2A - ChIP Grade
Host species	Rabbit
Tested applications	**Suitable for:** IHC-P, ELISA, WB, ChIP
Species reactivity	**Reacts with:** Human, Caenorhabditis elegans, Drosophila melanogaster, Schizosaccharomyces pombe **Predicted to work with:** Mouse, Rat, Chicken, Saccharomyces cerevisiae, Xenopus laevis
Immunogen	Synthetic peptide corresponding to Human Histone H2A aa 1-15. This antibody was generated by immunizing rabbits with a mixture of synthetic peptides containing amino acid residues 1-15 (MSGRGKQGGKARAKA) and 81-96 (PRHLQLAIRNDEELNK) of human Histone H2A. Sequence: PRHLQLAIRNDEELNK (1-15), PRHLQLAIRNDEELNK (81-96) Run BLAST with Run BLAST with
Positive control	Human peripheral blood cell lysates or Jurkat cell lysate.

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	Preservative: 0.02% Sodium azide Constituents: 99% PBS, 0.05% BSA
Purity	Protein G purified
Purification notes	Protein G Chromatography was used to purify this Histone H2A antibody
Clonality	Polyclonal
Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab13923 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
IHC-P		Use a concentration of 5 µg/ml.
ELISA		1/1000. See Dai et al, 2008 for details on Nucleosome ELISA
WB	★★★★★ (3)	Use a concentration of 2 µg/ml. Detects a band of approximately 14 kDa (predicted molecular weight: 16 kDa).
ChIP		Use at an assay dependent concentration. PubMed: 20711347

Target

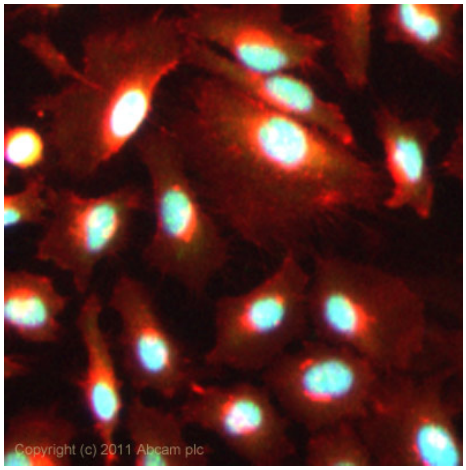
Function Core component of nucleosome. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA 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 H2A family.

Post-translational modifications The chromatin-associated form is phosphorylated on Thr-121 during mitosis.
Deiminated on Arg-4 in granulocytes upon calcium entry.
Monoubiquitination of Lys-120 by RING1 and RNF2/RING2 complex gives a specific tag for epigenetic transcriptional repression and participates in X chromosome inactivation of female mammals. It is involved in the initiation of both imprinted and random X inactivation. Ubiquitinated H2A is enriched in inactive X chromosome chromatin. Ubiquitination of H2A functions downstream of methylation of 'Lys-27' of histone H3. Monoubiquitination of Lys-120 by RNF2/RING2 can also be induced by ultraviolet and may be involved in DNA repair. Following DNA double-strand breaks (DSBs), it is ubiquitinated through 'Lys-63' linkage of ubiquitin moieties by the E2 ligase UBE2N and the E3 ligases RNF8 and RNF168, leading to the recruitment of repair proteins to sites of DNA damage. Monoubiquitination and ionizing radiation-induced 'Lys-63'-linked ubiquitination are distinct events.
Phosphorylation on Ser-2 is enhanced during mitosis. Phosphorylation on Ser-2 by RPS6KA5/MSK1 directly represses transcription. Acetylation of H3 inhibits Ser-2 phosphorylation by RPS6KA5/MSK1.
Symmetric dimethylation on Arg-4 by the PRDM1/PRMT5 complex may play a crucial role in the germ-cell lineage.

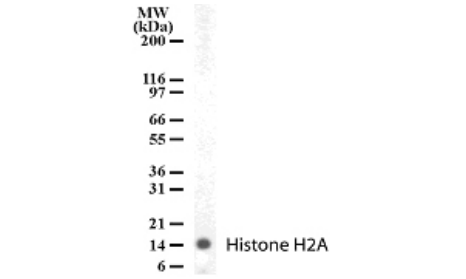
Cellular localization Nucleus. Chromosome.

Images



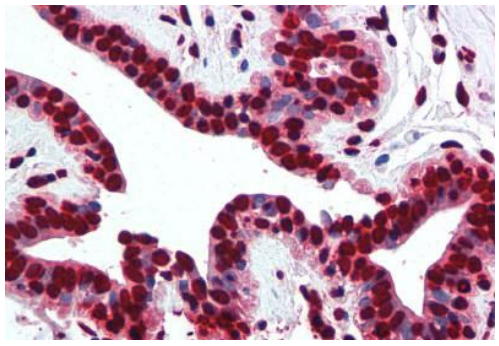
Immunocytochemistry/ Immunofluorescence - Anti-Histone H2A antibody - ChIP Grade (ab13923)

ICC/IF image of ab13923 stained HeLa cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody (ab13923, 1µg/ml) overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti-rabbit IgG (H+L) ab150077) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43µM.



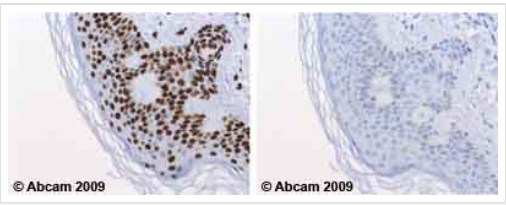
Western blot - Anti-Histone H2A antibody - ChIP Grade (ab13923)

Western blot analysis of Histone H2A in human PBMC cell lysate with ab13923.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A antibody - ChIP Grade (ab13923)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human breast tissue (ab30090)labelling Histone H2A with ab13923 at 5µg/ml. Staining was enhanced by boiling tissue sections in 10mM sodium citrate buffer, pH6.0 for 10-20 minutes followed by cooling at room temperature for 20 minutes.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2A antibody - ChIP Grade (ab13923)

ab13923 staining Human normal skin (ab30166). Staining is localised to nuclear compartment.
Left panel: with primary antibody at 1 ug/ml. Right panel: isotype control.
Sections were stained using an automated system DAKO Autostainer Plus , at room temperature: sections were rehydrated and antigen retrieved with the Dako 3 in 1 AR buffers citrate pH6.1 in a DAKO PT Link. Slides were peroxidase blocked in 3% H2O2 in methanol for 10 mins. They were then blocked with Dako Protein block for 10 minutes (containing casein 0.25% in PBS) then incubated with primary antibody for 20 min and detected with Dako envision flex amplification kit for 30 minutes. Colorimetric detection was completed with Diaminobenzidine for 5 minutes. Slides were counterstained with Haematoxylin and coverslipped under DePeX. Please note that for manual staining we recommend to optimize the primary antibody concentration and incubation time (overnight incubation), and amplification may be required.

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