abcam

Product datasheet

Anti-Histone H2B antibody - ChIP Grade ab1790

28 Abreviews 233 References 9 Images

Overview

Product name	Anti-Histone H2B antibody - ChIP Grade
Description	Rabbit polyclonal to Histone H2B - ChIP Grade
Host species	Rabbit
Specificity	This antibody is specific for Histone 2B.
Tested applications	**Suitable for:** WB, IHC-P, ChIP, ICC/IF, IP
Species reactivity	**Reacts with:** Cow, Human, Saccharomyces cerevisiae, Xenopus laevis, Arabidopsis thaliana **Predicted to work with:** Mouse, Rat, Chicken, Zebrafish
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers. (Peptide available as **ab16101**)
Positive control	Calf Thymus Histone Preparation This antibody gave a positive result when used in the following methanol fixed cell lines: HeLa
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
Light chain type	unknown

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab1790 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	★★★★★ (18)	Use a concentration of 0.1 µg/ml. Detects a band of approximately 17 kDa (predicted molecular weight: 14 kDa).Can be blocked with **Human Histone H2B peptide (ab16101)**.
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval before commencing with IHC staining protocol.
ChIP	★★★★☆ (3)	Use a concentration of 2 - 3 µg/ml.
ICC/IF	★★★★☆ (2)	Use a concentration of 0.5 µg/ml.
IP	★★★★★ (3)	Use a concentration of 5 µg/ml.

Target

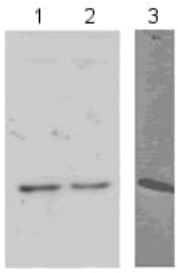
Relevance

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

Images



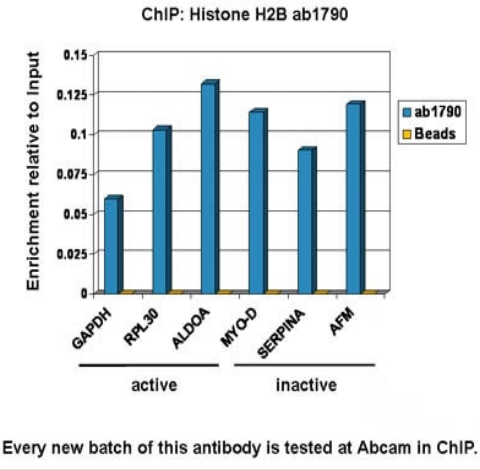
Western blot - Anti-Histone H2B antibody - ChIP Grade (ab1790)

All lanes : Anti-Histone H2B antibody - ChIP Grade (ab1790)

Lane 1 : Hela Histone prep
Lane 2 : Hela whole cell lysate
Lane 3 : S. cerevisiae whole cell lysate

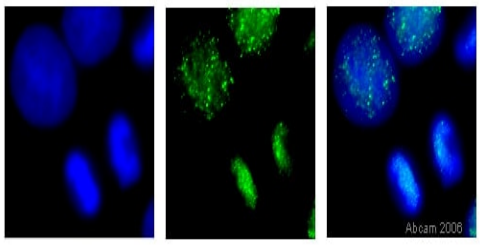
Performed under reducing conditions.

Predicted band size: 14 kDa



ChIP - Anti-Histone H2B antibody - ChIP Grade (ab1790)

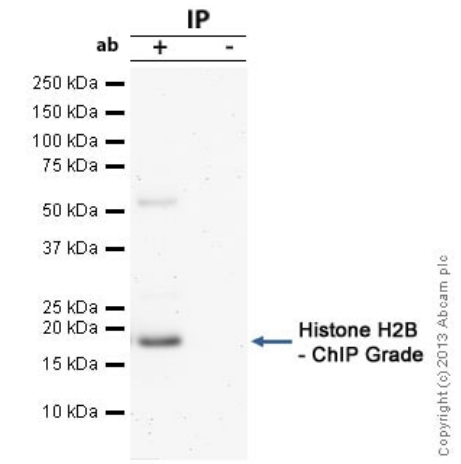
Chromatin was prepared from U2OS cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 2µg of ab1790 (blue), and 20µl of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci). Primers and probes are located in the first kb of the transcribed region.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2B antibody - ChIP Grade (ab1790)

This image is courtesy of Roberto Giambruno and Patrizia Lavia

HeLa cells were fixed in 100% methanol for 6 minutes at -20°C. The cells were washed 3 times in PBS then incubated with ab1790 (0.5µg/ml) for 1 hour at room temperature. The panel of images shows the cells stained with ab1790 (green) and counterstained with DAPI (blue). 100x magnification.



Immunoprecipitation - Anti-Histone H2B antibody - ChIP Grade (ab1790)

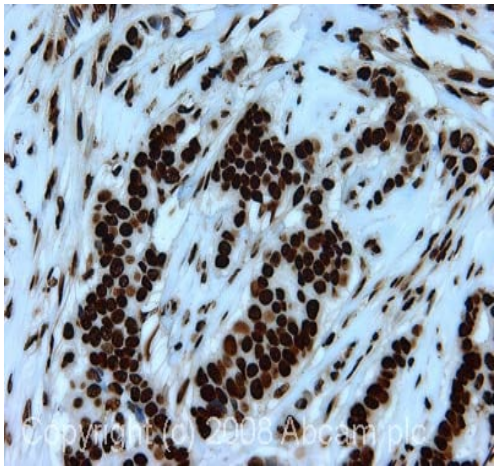
Histone H2B - ChIP Grade was immunoprecipitated using 0.5mg HeLa whole cell extract, 5µg of Rabbit polyclonal to and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, HeLa whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab1790.

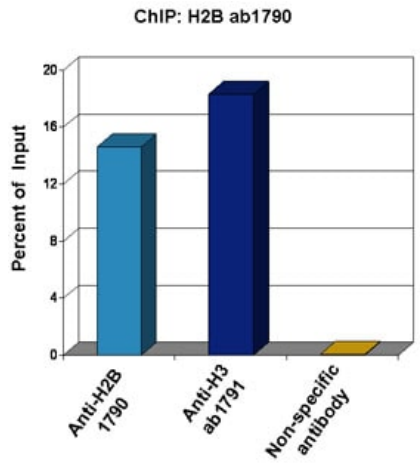
Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (**ab99697**).

Band: 14kDa; Histone H2B - ChIP Grade



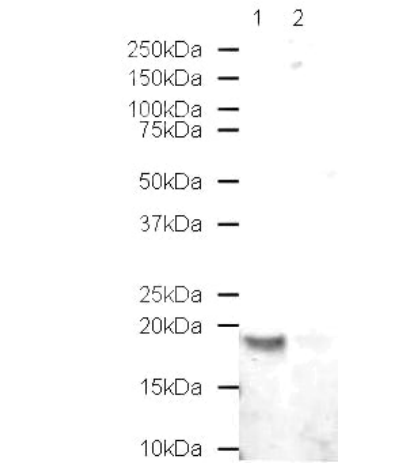
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2B antibody - ChIP Grade (ab1790)

IHC image of Histone H2B staining in human breast carcinoma FFPE section, performed on a Bond^{TM} system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab1790, 1µg/ml, for 8 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.



ChIP - Anti-Histone H2B antibody - ChIP Grade (ab1790)

Chromatin from Xenopus laevis oocytes was prepared according to the Abcam X-ChIP protocol. Oocytes were fixed with formaldehyde for 10 min. The ChIP was performed with 25 µg of chromatin, 3 µg of ab1790 (anti-H2B, light blue) and 3 µg of **ab1791** (anti-H3, dark blue), and 20 µl of Protein A/G sepharose beads. A non-specific antibody was used as a control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach).



Western blot - Anti-Histone H2B antibody - ChIP Grade (ab1790)

All lanes : Anti-Histone H2B antibody - ChIP Grade (ab1790) at 0.1 µg/ml

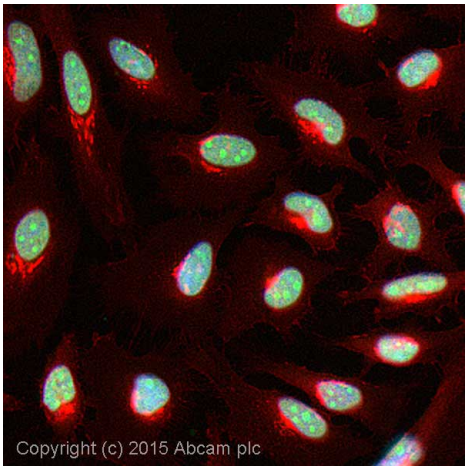
Lane 1 : Calf thymus histone prep
Lane 2 : Calf thymus histone prep with Human Histone H2B peptide (**ab16101**) at 1 µg/ml

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Alexa fluor Goat polyclonal anti-Rabbit IgG at 1/10000 dilution

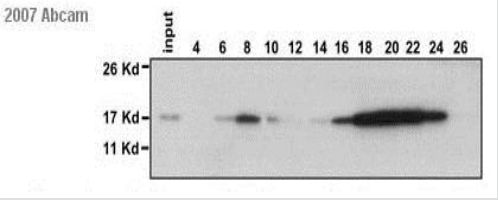
Performed under reducing conditions.

Predicted band size: 14 kDa



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2B antibody - ChIP Grade (ab1790)

ICC/IF image of ab1790 stained HeLa cells. The cells were 100% methanol fixed (5 min) then permeabilised using 0.1% PBS-Triton and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to further permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab1790 at 0.1µg/ml overnight at +4°C. The secondary antibody (pseudo-colored green) was Alexa Fluor® 488 goat anti- rabbit (**ab150081**) IgG (H+L) preadsorbed, used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (pseudo-colored red) at a 1/200 dilution for 1h at room temperature. DAPI was used to stain the cell nuclei (pseudo-colored blue) at a concentration of 1.43µM for 1hour at room temperature.



Western blot - Anti-Histone H2B antibody - ChIP Grade (ab1790)

This image is courtesy of an Abreview submitted by Dr Igor Landais

ab1790 at 1/3000 detecting Histone H2B from Xenopus laevis (S phase egg extracts - whole cell lysates 60ug per lane) by Western Blot. The egg extracts were fractionated using a gel filtration column and every other fraction (4 - 26) was loaded onto a 8-16% gel. The input corresponds to 1ul of crude extract. In this experiment an HRP conjugated donkey anti-rabbit antibody was used as the secondary.

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