

Product datasheet

Anti-Histone H2B (crotonyl K5) antibody [EPR17483] ab177396

Recombinant RabMAb

8 Images

Overview

Product name	Anti-Histone H2B (crotonyl K5) antibody [EPR17483]
Description	Rabbit monoclonal [EPR17483] to Histone H2B (crotonyl K5)
Host species	Rabbit
Tested applications	Suitable for: PepArr, IHC-P, WB, ICC/IF
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HeLa and NIH3T3 cell lysates;. IHC: Human colon, mouse colon, rat liver tissues. ICC/IF: HeLa cells.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.</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	Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR17483
Isotype	IgG

Applications

The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab177396 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
PepArr		Use at an assay dependent concentration.
IHC-P		1/500. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/500. Detects a band of approximately 14 kDa (predicted molecular weight: 14 kDa).
ICC/IF		1/250.

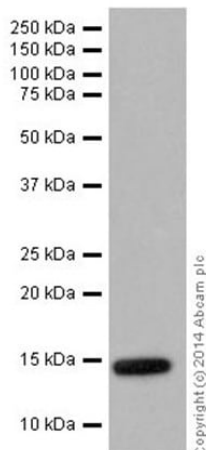
Target

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



Western blot - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)
at 1/1000 dilution + HeLa (Human epithelial cells from cervix adenocarcinoma) cell lysate at 20 µg

Secondary

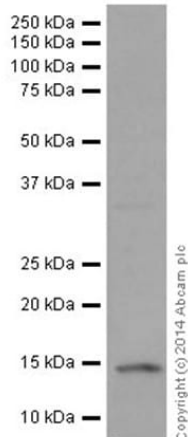
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 14 kDa

Observed band size: 14 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDm/TBST.



Western blot - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)
at 1/500 dilution + NIH/3T3 (Mouse embryo fibroblast cells) cell lysate at 10 µg

Secondary

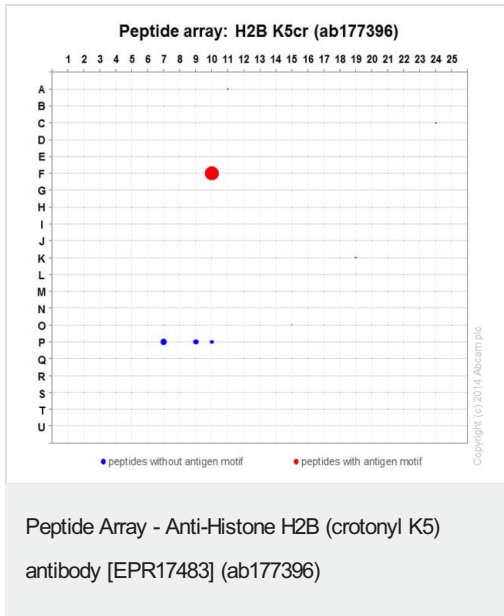
Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 14 kDa

Observed band size: 14 kDa

Exposure time: 3 minutes

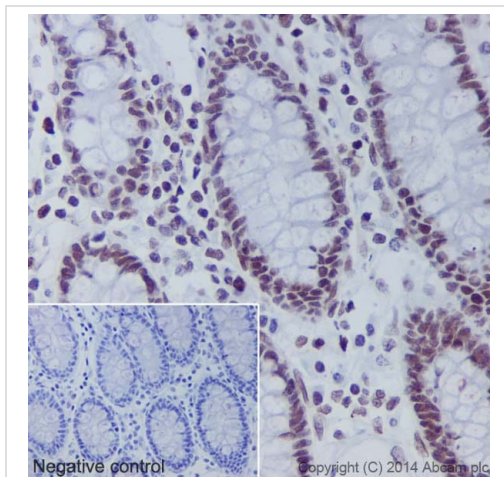
Blocking/Dilution buffer: 5% NFDm/TBST.



ab177396 was tested in Peptide Array against 501 different modified and unmodified histone peptides; each peptide is printed on the array at six concentrations (each in triplicate).

Circle area represents affinity between the antibody and a peptide: all antigen-containing peptides are displayed as red circles, all other peptides as blue circles. The affinity is calculated as area under curve when antibody binding values are plotted against the corresponding peptide concentration. Each circle area is normalized to the peptide with the strongest affinity.

The complete dataset, including full list of all peptides and information on the position of each peptide in the diagram, can be downloaded [here](#).

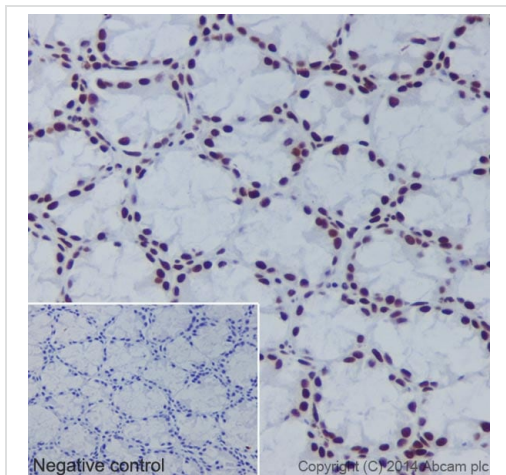


Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Histone H2B (crotonyl K5) using ab177396 at 1/500 dilution. A Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) was used as secondary at 1/500 dilution. Counter stained with Hematoxylin. Inset image: negative control obtained using PBS instead of ab177396.

Note: Nucleus staining on glandular epithelium of Human colon tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

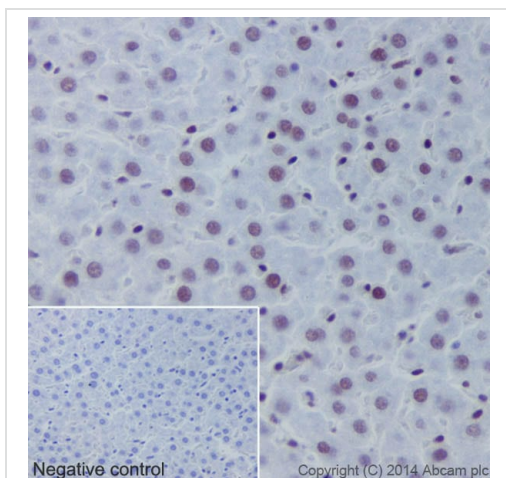


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

Immunohistochemical analysis of paraffin-embedded Mouse colon tissue labeling Histone H2B (crotonyl K5) using ab177396 at 1/500 dilution. A Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) was used as secondary at 1/500 dilution. Counter stained with Hematoxylin. Inset image: negative control obtained using PBS instead of ab177396.

Note: Nucleus staining on glandular epithelium of Mouse colon tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

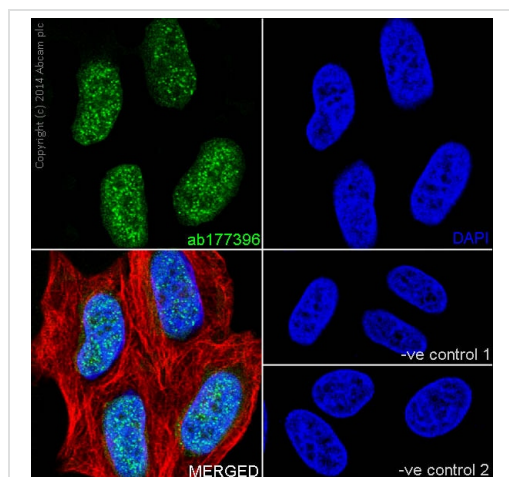


Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

Immunohistochemical analysis of paraffin-embedded Rat liver tissue labeling Histone H2B (crotonyl K5) using ab177396 at 1/500 dilution. A Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) was used as secondary at 1/500 dilution. Counter stained with Hematoxylin. Inset image: negative control obtained using PBS instead of ab177396.

Note: Nucleus staining on Rat liver tissue was observed.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling Histone H2B (crotonyl K5) with ab177396 at 1/250 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (**ab150077**) secondary antibody at 1/500 dilution (green). Confocal image showing nuclear staining on HeLa cell line. The nuclear counter stain is DAPI (blue). Tubulin is detected with **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution and **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

1. ab177396 at 1/250 dilution followed by **ab150120** (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution.
2. **ab7291** (anti-Tubulin mouse mAb) at 1/1000 dilution followed by **ab150077** (Alexa Fluor®488 Goat Anti-Rabbit IgG H&L) at 1/500 dilution.

Why choose a recombinant antibody?

Research with confidence
Consistent and reproducible results

Long-term and scalable supply
Recombinant technology

Success from the first experiment
Confirmed specificity

Ethical standards compliant
Animal-free production

Anti-Histone H2B (crotonyl K5) antibody [EPR17483] (ab177396)

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