# abcam

## Product datasheet

## Anti-Histone H2B antibody [EP819Y] - ChIP Grade ab52985

RabMAb

★★★★★ 2 Abreviews 1 References 4 Images

## Overview

**Product name** Anti-Histone H2B antibody [EP819Y] - ChIP Grade

**Description** Rabbit monoclonal [EP819Y] to Histone H2B - ChIP Grade

**Host species** Rabbit

**Tested applications** Suitable for: WB, IHC-P, ChIP

Unsuitable for: Flow Cyt or ICC/IF

Species reactivity Reacts with: Human

Predicted to work with: Mouse

**Immunogen** Synthetic peptide within Human Histone H2B. The exact sequence is proprietary.

Positive control ChIP: Chromatin prepared from HeLa cells. IHC-P: Human breast carcinoma and colon tissue.

WB: A431 cell lysate.

**General notes** Our RabMAb® technology is a patented hybridoma-based technology for making rabbit

monoclonal antibodies. For details on our patents, please refer to **RabMAb**® **patents**.

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

Rat: We have preliminary internal testing data to indicate this antibody may not react with this

species. Please contact us for more information.

## **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.

Avoid freeze / thaw cycle.

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Storage buffer pH: 7.20

Preservative: 0.05% Sodium azide

Constituents: 99% Tissue culture supernatant, 0.1% BSA

Purity Protein A purified

Clonality Monoclonal
Clone number EP819Y

**Isotype** IgG

#### **Applications**

## The Abpromise guarantee

Our **Abpromise guarantee** covers the use of ab52985 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	★★★☆☆(1)	1/10000. Detects a band of approximately 14 kDa (predicted molecular weight: 14 kDa).
IHC-P		1/1000. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ChIP	<b>★★★★</b> <u>(1)</u>	Use 2 µg for 25 µg of chromatin.

**Application notes** 

Is unsuitable for Flow Cyt or ICC/IF.

## **Target**

#### Relevance

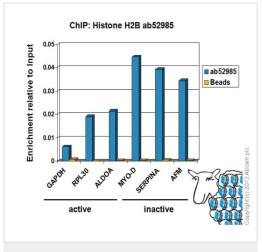
Core component of nucleosome. Nucleosomes wrap and compact DNA into chromatin, limiting DNA accessibility to the cellular machineries which require DNA as a template. Histones thereby play a central role in transcription regulation, DNA repair, DNA replication and chromosomal stability. DNA accessibility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

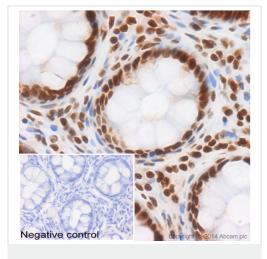
Nuclear

#### **Images**



ChIP - Anti-Histone H2B antibody [EP819Y] - ChIP Grade (ab52985)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 5µg of ab52985 (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). Primers and probes are located in the first kb of the transcribed region.

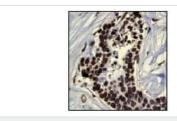


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2B antibody

[EP819Y] - ChIP Grade (ab52985)

IHC image of ab52985 staining Histone H2B in human colon formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. 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 ab52985, 1/1000 dilution, for 15 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. No primary antibody was used in the negative control (shown on the inset).

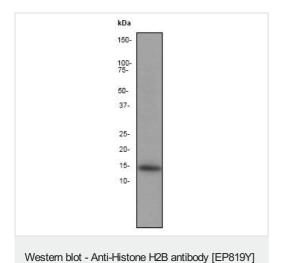
For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2B antibody
[EP819Y] - ChIP Grade (ab52985)

ab52985, at a 1/100 dilution, staining Human Histone H2B in breast carcinoma using Immunohistochemistry, Paraffin Embedded tissue.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



- ChIP Grade (ab52985)

Anti-Histone H2B antibody [EP819Y] - ChIP Grade (ab52985) at 1/10000 dilution + A431 cell lysate at 10 µg

## Secondary

goat anti-rabbit HRP labelled at 1/2000 dilution

**Predicted band size:** 14 kDa **Observed band size:** 14 kDa

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

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- We provide support in Chinese, English, French, German, Japanese and Spanish
- Extensive multi-media technical resources to help you
- We investigate all quality concerns to ensure our products perform to the highest standards

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