## abcam

### Product datasheet

# HRP Anti-Histone H3 antibody - Nuclear Loading Control ab21054

**29 References** 3 Images

Overview

Product name HRP Anti-Histone H3 antibody - Nuclear Loading Control

**Description** HRP Rabbit polyclonal to Histone H3 - Nuclear Loading Control

Host species Rabbit

Conjugation HRP

Tested applications Suitable for: WB, IHC-P

Species reactivity Reacts with: Mouse, Human, Saccharomyces cerevisiae, Drosophila melanogaster

Immunogen Synthetic peptide corresponding to Human Histone H3 aa 100 to the C-terminus conjugated to

keyhole limpet haemocyanin.

Database link: P68431

(Peptide available as ab12149)

Positive control This antibody gave a positive signal in the following: HeLa (Human epithelial carcinoma cell line)

Whole Cell Lysate NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate Yeast S.cerevisiae Whole Cell Lysate Drosophila Embryo Nuclear Extract IHC-P: FFPE human skin

(normal) and human cervix (normal) tissue sections.

**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). Store at -20°C. Avoid freeze / thaw cycle.

Store In the Dark.

Storage buffer pH: 7.40

Preservative: 0.1% 10% Proclin 300 Solution Constituents: 30% Glycerol, 1% BSA, PBS

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Purity Immunogen affinity purified

**Clonality** Polyclonal

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab21054 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		1/5000. Detects a band of approximately 17 kDa (predicted molecular weight: 15.4 kDa).
IHC-P		Use a concentration of 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

#### **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 H3 family.

**Developmental stage** 

Expressed during S phase, then expression strongly decreases as cell division slows down

during the process of differentiation.

Post-translational modifications

Acetylation is generally linked to gene activation. Acetylation on Lys-10 (H3K9ac) impairs methylation at Arg-9 (H3R8me2s). Acetylation on Lys-19 (H3K18ac) and Lys-24 (H3K24ac) favors methylation at Arg-18 (H3R17me).

Citrullination at Arg-9 (H3R8ci) and/or Arg-18 (H3R17ci) by PAD4 impairs methylation and represses transcription.

Asymmetric dimethylation at Arg-18 (H3R17me2a) by CARM1 is linked to gene activation. Symmetric dimethylation at Arg-9 (H3R8me2s) by PRMT5 is linked to gene repression. Asymmetric dimethylation at Arg-3 (H3R2me2a) by PRMT6 is linked to gene repression and is mutually exclusive with H3 Lys-5 methylation (H3K4me2 and H3K4me3). H3R2me2a is present at the 3' of genes regardless of their transcription state and is enriched on inactive promoters, while it is absent on active promoters.

Methylation at Lys-5 (H3K4me), Lys-37 (H3K36me) and Lys-80 (H3K79me) are linked to gene activation. Methylation at Lys-5 (H3K4me) facilitates subsequent acetylation of H3 and H4. Methylation at Lys-80 (H3K79me) is associated with DNA double-strand break (DSB) responses and is a specific target for TP53BP1. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are linked to gene repression. Methylation at Lys-10 (H3K9me) is a specific target for HP1 proteins (CBX1, CBX3 and CBX5) and prevents subsequent phosphorylation at Ser-11 (H3S10ph) and acetylation of H3 and H4. Methylation at Lys-5 (H3K4me) and Lys-80 (H3K79me) require preliminary monoubiquitination of H2B at 'Lys-120'. Methylation at Lys-10 (H3K9me) and Lys-28 (H3K27me) are enriched in inactive X chromosome chromatin.

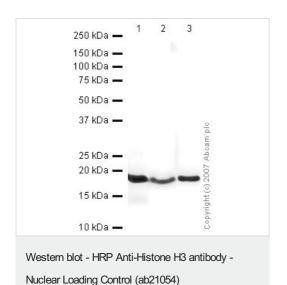
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and RPS6KA5 is important during interphase because it enables the transcription of genes following external stimulation, like mitogens, stress, growth factors or UV irradiation and result in the activation of genes, such as c-fos and c-jun. Phosphorylation at Ser-11 (H3S10ph), which is linked to gene activation, prevents methylation at Lys-10 (H3K9me) but facilitates acetylation of H3 and H4. Phosphorylation at Ser-11 (H3S10ph) by AURKB mediates the dissociation of HP1 proteins (CBX1, CBX3 and CBX5) from heterochromatin. Phosphorylation at Ser-11 (H3S10ph) is also an essential regulatory mechanism for neoplastic cell transformation. Phosphorylated at Ser-29 (H3S28ph) by MLTK isoform 1, RPS6KA5 or AURKB during mitosis or upon ultraviolet B irradiation. Phosphorylation at Thr-7 (H3T6ph) by PRKCBB is a specific tag for epigenetic transcriptional activation that prevents demethylation of Lys-5 (H3K4me) by LSD1/KDM1A. At centromeres, specifically phosphorylated at Thr-12 (H3T11ph) from prophase to early anaphase, by DAPK3 and PKN1. Phosphorylation at Thr-12 (H3T11ph) by PKN1 is a specific tag for epigenetic transcriptional activation that promotes demethylation of Lys-10 (H3K9me) by KDM4C/JMJD2C. Phosphorylation at Tyr-42 (H3Y41ph) by JAK2 promotes exclusion of CBX5 (HP1 alpha) from chromatin.

Monoubiquitinated by RAG1 in lymphoid cells, monoubiquitination is required for V(D)J recombination (By similarity). 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.

Nucleus. Chromosome.

#### **Cellular localization**

**Images** 



All lanes: HRP Anti-Histone H3 antibody - Nuclear Loading

Control (ab21054) at 1/5000 dilution

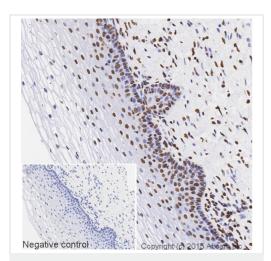
**Lane 1**: NIH/3T3 whole cell lysate (<u>ab7179</u>) at 10 μg

Lane 2 : Yeast S.cerevisiae Whole Cell Lysate at 10  $\mu g$ 

Lane 3: Drosophila Embryo Nuclear Extract at 20 μg

Performed under reducing conditions.

**Predicted band size:** 15.4 kDa **Observed band size:** 17 kDa

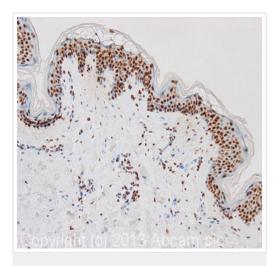


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - HRP Anti-Histone H3 antibody - Nuclear Loading Control (ab21054)

IHC image of Histone H3 staining in a section of formalin-fixed paraffin-embedded normal human cervix tissue\*, performed on a Leica BOND<sup>TM</sup>. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20mins. The section was then incubated with ab21054, 1/500 dilution, for 15 mins at room temperature. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. The inset negative control image is taken from an identical assay without primary antibody.

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.

\*Tissue obtained from the Human Research Tissue Bank, supported by the NIHR Cambridge Biomedical Research Centre



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - HRP Anti-Histone H3 antibody - Nuclear Loading Control (ab21054)

IHC image of Histone H3 staining in human skin formalin fixed paraffin embedded tissue section, performed on a Leica Bond 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 ab21054, 1µg/ml, 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.

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.

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