# abcam

# Product datasheet

# Anti-Histone H3.3 antibody ab213677

### 5 Images

#### Overview

**Product name** Anti-Histone H3.3 antibody

**Description** Rabbit polyclonal to Histone H3.3

Host species Rabbit

**Tested applications** Suitable for: Flow Cyt (Intra), IHC-P, WB

Species reactivity Reacts with: Mouse, Rat, Human

Predicted to work with: Pig ...

Immunogen Synthetic peptide within Human Histone H3.3 aa 50 to the C-terminus conjugated to keyhole

limpet haemocyanin. The exact immunogen sequence used to generate this antibody is proprietary information. If additional detail on the immunogen is needed to determine the suitability of the antibody for your needs, please **contact** our Scientific Support team to discuss

your requirements.

Database link: P84243

Run BLAST with
Run BLAST with

Positive control WB: K562 cell lysate. Flow Cyt (Intra): Mouse splenocytes. IHC-P: Rat brain, mouse intestine and

human colon cancer tissues.

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 long

term. Avoid freeze / thaw cycle.

Storage buffer pH: 7.40

Preservative: 0.02% Proclin 300

Constituents: 50% Glycerol (glycerin, glycerine), 1% BSA, 48.98% TBS, 1X

1

Purity Protein A purified

**Clonality** Polyclonal

**Isotype** IgG

#### **Applications**

#### The Abpromise guarantee

Our Abpromise guarantee covers the use of ab213677 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
Flow Cyt (Intra)		1/20 - 1/100.
IHC-P		1/100 - 1/500. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol. When using a fluorescent probe the recommended dilution is 1/50 – 1/200.
WB		1/100 - 1/1000. Predicted molecular weight: 15 kDa.

#### **Target**

#### **Function**

Variant histone H3 which replaces conventional H3 in a wide range of nucleosomes in active genes. Constitutes the predominant form of histone H3 in non-dividing cells and is incorporated into chromatin independently of DNA synthesis. Deposited at sites of nucleosomal displacement throughout transcribed genes, suggesting that it represents an epigenetic imprint of transcriptionally active chromatin. 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

**Developmental stage** 

Post-translational modifications

Belongs to the histone H3 family.

Expressed throughout the cell cycle independently of DNA synthesis.

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.

Specifically enriched in modifications associated with active chromatin such as methylation at Lys-5 (H3K4me), Lys-37 and Lys-80. 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), which are linked to gene repression, are underrepresented.

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.

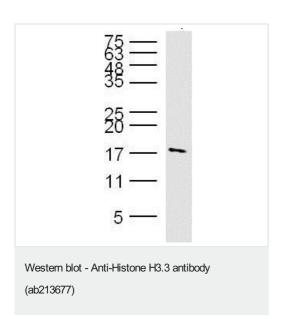
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and 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. Phosphorylation on Ser-32 (H3S31ph) is specific to regions bordering centromeres in metaphase chromosomes.

**Cellular localization** 

Nucleus. Chromosome.

V(D)J recombination.

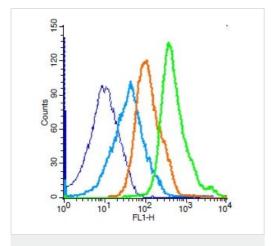
#### **Images**



Anti-Histone H3.3 antibody (ab213677) at 1/300 dilution + K562 cell lysate

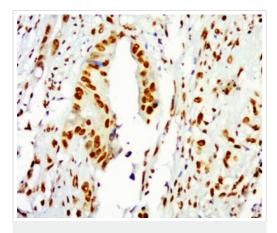
Ubiquitinated. Monoubiquitinated by RAG1 in lymphoid cells, monoubiquitination is required for

Predicted band size: 15 kDa



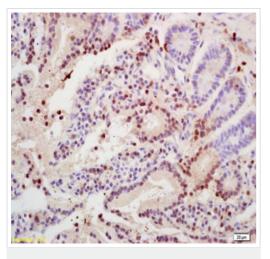
Flow Cytometry (Intracellular) - Anti-Histone H3.3 antibody (ab213677)

Intracellular flow cytometric analysis ofMouse splenocytes labeling Histone H3.3 with ab213677 at 1/50 dilution for 30 minutes (green)followed by a FITC conjugated secondary antibody compared to unstained cells (blue), secondary only (light blue), and isotype control (orange).



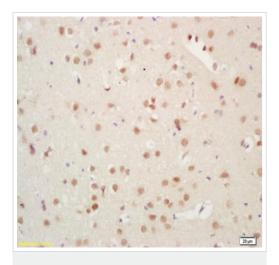
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3.3 antibody (ab213677)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded human colon cancer tissue labeling Histone H3.3 with ab213677 at 1/400 dilution followed by conjugation to the secondary antibody and DAB staining.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3.3 antibody (ab213677)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded Mouse intestine tissue labeling Histone H3.3 with ab213677 at 1/200 dilution followed by conjugation to the secondary antibody and DAB staining.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3.3 antibody (ab213677)

Immunohistochemical analysis of formalin-fixed, paraffin-embedded Rat brain tissue labeling Histone H3.3 with ab213677 at 1/200 dilution followed by conjugation to the secondary antibody and DAB staining.

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