abcam

Product datasheet

Anti-Histone H3 (methyl K37) antibody [EPR20970] ab215728

Recombinant RabMAb

8 Images

Overview

Product name	Anti-Histone H3 (methyl K37) antibody [EPR20970]
Description	Rabbit monoclonal [EPR20970] to Histone H3 (methyl K37)
Host species	Rabbit
Tested applications	**Suitable for:** PepArr, WB, Flow Cyt, ICC
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Histone H3 aa 1-100 (methyl K37). The exact sequence is proprietary. Database link: P68431
Positive control	WB: Recombinant mutant H3 chemically modified to mimic H3K37Me; NCCIT, HeLa, NIH/3T3 and PC-12 whole cell lysates. ICC: HeLa and NIH/3T3 cells. Flow cyt: HeLa and NIH/3T3 cells.
General notes	This product is a recombinant monoclonal antibody, which offers several advantages including: - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production For more information see here. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

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: 59.94% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal

Clone number	EPR20970
Isotype	IgG

Applications

Our Abpromise guarantee covers the use of **ab215728** 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 a concentration of 0.1 µg/ml.
WB		1/1000. Predicted molecular weight: 15 kDa.
Flow Cyt		1/60.
ICC		1/100.

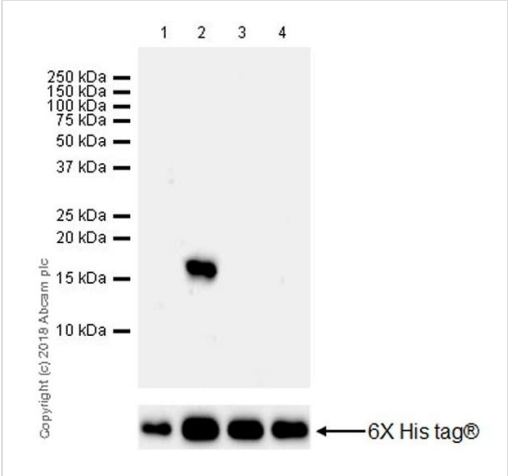
Target

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 PADI4 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.

Cellular localization Nucleus. Chromosome.

Images



Western blot - Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

All lanes : Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728) at 1/1000 dilution

Lane 1 : Unmodified recombinant H3 10 ng
Lane 2 : Recombinant mutant H3 chemically modified to mimic H3K37Me, 10ng
Lane 3 : Recombinant mutant H3 chemically modified to mimic H3K37Me2 10 ng
Lane 4 : Recombinant mutant H3 chemically modified to mimic H3K37Me3 10 ng

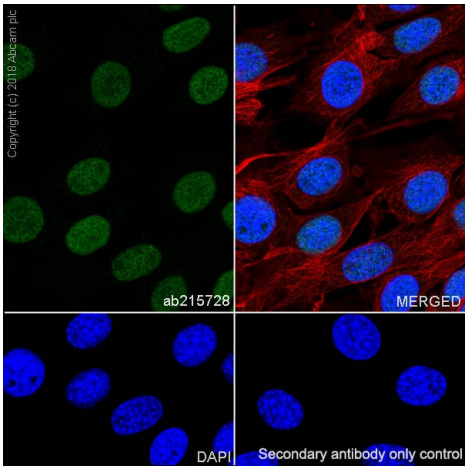
Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 15 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

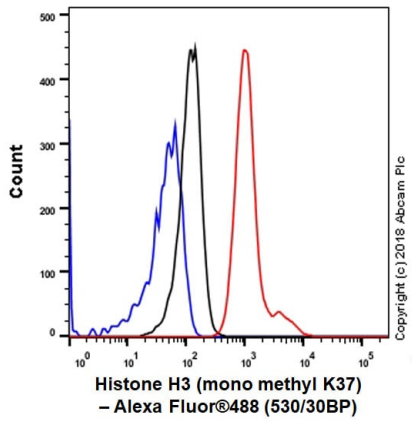
The recombinant proteins were His-tagged.



Immunocytochemistry - Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

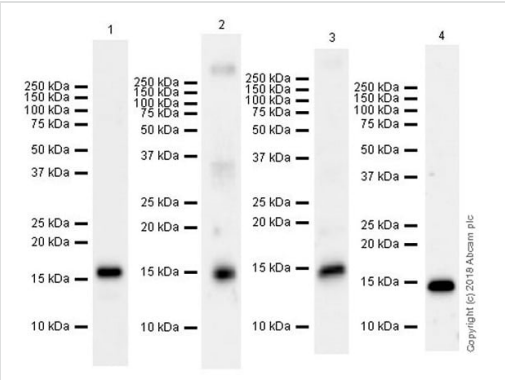
Immunofluorescent analysis of 4% paraformaldehyde-fixed 0.1% Triton X-100 permeabilized NIH/3T3 (mouse embryonic fibroblast cell line) cells labeling Histone H3 (methyl K37) with ab215728 at 1/100 dilution, followed by ab150077 AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining in NIH/3T3 cell line is observed. DAPI was used as the nuclear counterstain. Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) was used as the counterstain antibody at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is ab150077 AlexaFluor®488 Goat anti-Rabbit secondary at 1/1000 dilution.



Flow Cytometry - Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

Flow cytometric analysis of 90% methanol permeabilized, 4% paraformaldehyde-fixed NIH/3T3 (mouse embryo fibroblast cell line) cells labeling Histone H3 (methyl K37) with ab215728 at 1/60 dilution (Red) compared with Rabbit monoclonal IgG (ab172730) (black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody) (blue). Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 was used as the secondary antibody.



Western blot - Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

All lanes : Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728) at 1/1000 dilution

Lane 1 : NCCIT (human pluripotent embryonic carcinoma epithelial cell) whole cell lysate
Lane 2 : HeLa (human cervix adenocarcinoma epithelial cell) whole cell lysate
Lane 3 : NIH/3T3 (mouse embryonic fibroblast) whole cell lysate
Lane 4 : PC-12 (rat adrenal gland pheochromocytoma) whole cell lysate

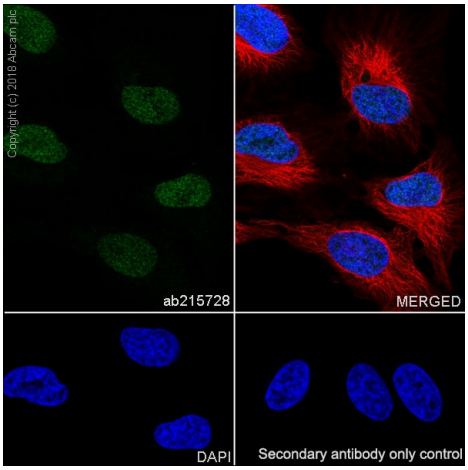
Lysates/proteins at 10 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 15 kDa

Exposure times:
Lane 1: 26 seconds;
Lane 2: 3 minutes;
Lane 3: 81 seconds;
Lane 4: 3 minutes.

Blocking/Dilution buffer: 5% BSA/TBST



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Immunofluorescent analysis of 4% paraformaldehyde-fixed 0.1% Triton X-100 permeabilized HeLa (human cervix adenocarcinoma epithelial cell line) cells labeling Histone H3 (methyl K37) with ab215728 at 1/100 dilution, followed by ab150077 AlexaFluor®488 Goat anti-Rabbit secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining in HeLa cell line is observed. DAPI was used as the nuclear counterstain. Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594) (ab195889) was used as the counterstain antibody at 1/200 dilution (red).

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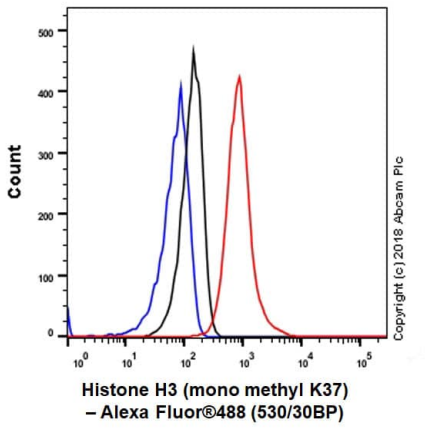

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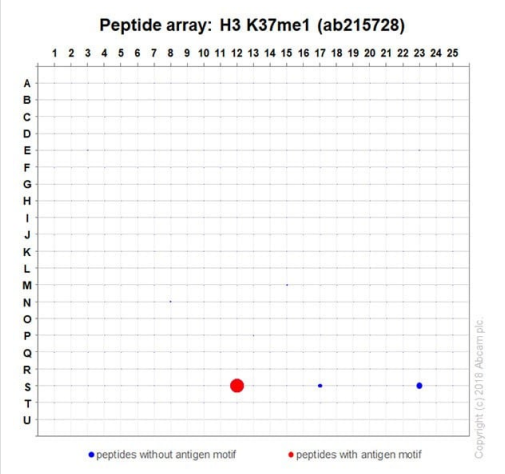

Peptide Array - Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

ab215728 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).

Blocking buffer: 5% BSA in TBST.

Primary antibody concentration: 0.1 µg/ml.

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.

Anti-Histone H3 (methyl K37) antibody [EPR20970] (ab215728)

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