abcam

Product datasheet

Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade ab177184

יילעבער RabMAb



★★★★★ 1 Abreviews 5 References

画像数 9

製品の概要

製品名 Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade

製品の詳細 Rabbit monoclonal [EPR17467] to Histone H3 (di methyl K79) - ChIP Grade

由来種 Rabbit

アプリケーション 適用あり: PepArr, ChIP, IHC-P, WB, ICC/IF

種交差性 交差種: Mouse, Rat, Human

免疫原 Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.

ポジティブ・コントロール WB: HeLa and NIH/3T3 cell lysates, IHC-P: Human colon, mouse liver and rat pancreas tissues.

ICC/IF: HeLa cells. ChIP: Chromatin prepared from HeLa cells.

特記事項 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.

製品の特性

製品の状態 Liquid

保存方法 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.

バッファー Preservative: 0.01% Sodium azide

Constituents: 59% PBS, 40% Glycerol, 0.05% BSA

精製度 Protein A purified

ポリモノ モノクローナル

クローン名 EPR17467

アイソタイプ ΙgG

The Abpromise guarantee <u>Abpromise保証は、</u>次のテスト済みアプリケーションにおけるab177184の使用に適用されます アプリケーションノートには、推奨の開始希釈率がありますが、適切な希釈率につきましてはご検討ください。

アプリケーション	Abreviews	特記事項
PepArr		Use at an assay dependent concentration.
ChIP		Use 5 µg for 25 µg of chromatin.
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB		1/2500. Detects a band of approximately 15 kDa (predicted molecular weight: 15 kDa).
ICC/IF	★音音音音 (1)	1/2000.

ターゲット情報

機能

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.

配列類似性 発生段階

Belongs to the histone H3 family.

翻訳後修飾

Expressed during S phase, then expression strongly decreases as cell division slows down during the process of differentiation.

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.

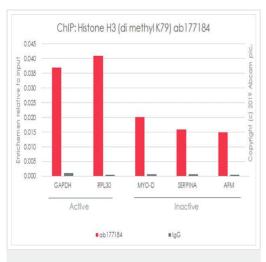
Phosphorylated at Thr-4 (H3T3ph) by GSG2/haspin during prophase and dephosphorylated during anaphase. Phosphorylation at Ser-11 (H3S10ph) by AURKB is crucial for chromosome condensation and cell-cycle progression during mitosis and meiosis. In addition phosphorylation at Ser-11 (H3S10ph) by RPS6KA4 and 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 Tvr-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.

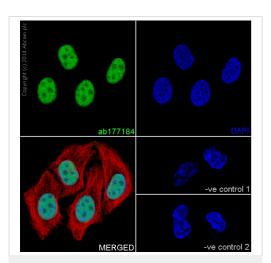
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画像

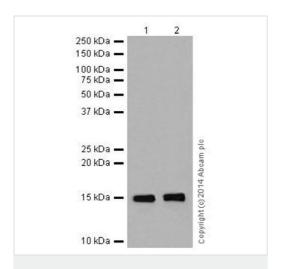


ChIP - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25 μg of chromatin, 5 μg of ab177184 (red), and 20 μl of Protein A/G Sepharose beads. 5 μg of rabbit normal IgG was added to the beads control (grey). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci). Primers and probes are located in the first kb of the transcribed region



Immunocytochemistry/ Immunofluorescence - Anti-Histone H3 (di methyl K79) antibody [EPR17467] -ChIP Grade (ab177184)



Western blot - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells labeling Histone H3 (di methyl K79) with ab177184 at 1/2000 dilution, followed by Goat anti-rabbit
Alexa Fluor® 488 (ab150077) secondary antibody at 1/500 dilution (green). Nuclear staining on HeLa cell line is observed. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (goat anti-mouse AlexaFluor®594 secondary) at 1/500 dilution (red).

The negative controls are as follows;

- 1. ab177184 at 1/2000 dilution followed by <u>ab150120</u> (goat <u>antimouse AlexaFluor®594</u> secondary) at 1/500 dilution.
- 2. <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (goat <u>anti-rabbit Alexa Fluor®488</u>; lgG H&L) at 1/400 dilution.

All lanes : Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184) at 1/2500 dilution

Lane 1: HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysate

Lane 2: NIH/3T3 (Mouse embyro fibroblast cells) whole cell lysate

Lysates/proteins at 20 µg per lane.

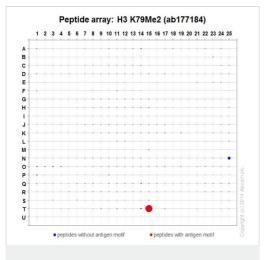
Secondary

All lanes : Goat Anti-Rabbit lgG, (H+L), Peroxidase conjugated at 1/1000 dilution

Predicted band size: 15 kDa **Observed band size:** 15 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

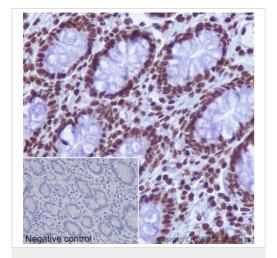


Peptide Array - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

ab177184 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).

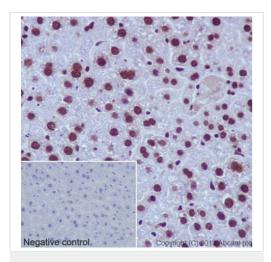
Circle area represents affinity between the antibody and a peptide: all antigen-containing peptides are displayed as red circles, all other peptides as blue circles. The affinity is calculated as area under curve when antibody binding values are plotted against the corresponding peptide concentration.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



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Histone H3 (di methyl K79) with ab177184 at 1/2000 dilution, followed by Goat **Anti-Rabbit HRP** (lgG H&L) at 1/500 dilution (**ab97051**). Nucleus staining on glandular epithelium of Human colon tissue is observed. Counter stained with Hematoxylin. Negative control: Using PBS instead of primary antibody. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

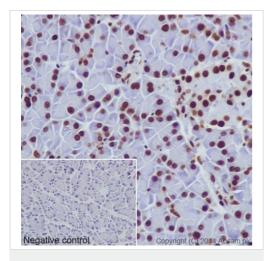


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

Immunohistochemical analysis of paraffin-embedded Mouse liver tissue labeling Histone H3 (di methyl K79) with ab177184 at 1/2000 dilution, followed by Goat **Anti-Rabbit HRP** (lgG HRP) at 1/500 dilution (**ab97051**). Nucleus staining mouse liver tissue is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody.

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

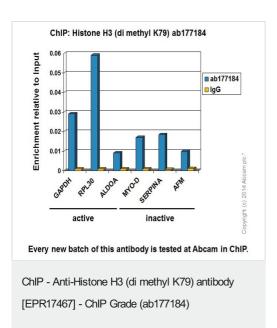


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H3 (di methyl K79) antibody [EPR17467] - ChIP Grade (ab177184)

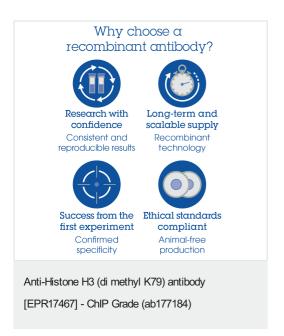
Immunohistochemical analysis of paraffin-embedded Rat pancreas tissue labeling Histone H3 (di methyl K79) with ab177184 at 1/2000 dilution, followed by Goat **Anti-Rabbit HRP** (lgG H&L) at 1/500 dilution (**ab97051**). Nucleus staining on rat pancreas tissue is observed. Counter stained with Hematoxylin.

Negative control: Using PBS instead of primary antibody.

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



Chromatin was prepared from Hela (Human epithelial cells from cervix adenocarcinoma) cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10min. The ChIP was performed with 25µg of chromatin, 10µg of ab177184 (blue), and 20µl of Anti rabbit IgG sepharose beads. 10µg of Rabbit normal IgG was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (SYBR approach). Primers and probes are located in the first kb of the transcribed region.



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