abcam

Product datasheet

Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade ab177312

יעלאעבע RabMAb

4 References 画像数 10

製品の概要

製品名 Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade

製品の詳細 Rabbit monoclonal [EPR17471] to Histone H2A (acetyl K9) - ChIP Grade

由来種 Rabbit

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

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 treated with 500 ng/ml Trichostatin A for 4 hours whole cell lysates. IHC-P:

Human colon, mouse liver and rat colon tissues. ICC/IF: HeLa cells. ChIP: Chromatin 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

クローン名

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.

Preservative: 0.01% Sodium azide バッファー

EPR17471

Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

精製度 Protein A purified

ポリモノ モノクローナル

アプリケーション

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ab177312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-sequencing | | Use at an assay dependent concentration. |
| WB | | 1/1000. Detects a band of approximately 14 kDa (predicted molecular weight: 14 kDa). |
| ICC/IF | | 1/2000. |
| IHC-P | | 1/4000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. |
| ChIP | | Use 2 µg for 25 µg of chromatin. |

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

翻訳後修飾

Belongs to the histone H2A family.

The chromatin-associated form is phosphorylated on Thr-121 during mitosis.

Deiminated on Arg-4 in granulocytes upon calcium entry.

Monoubiquitination of Lys-120 by RING1 and RNF2/RING2 complex gives a specific tag for epigenetic transcriptional repression and participates in X chromosome inactivation of female mammals. It is involved in the initiation of both imprinted and random X inactivation. Ubiquitinated H2A is enriched in inactive X chromosome chromatin. Ubiquitination of H2A functions downstream of methylation of 'Lys-27' of histone H3. Monoubiquitination of Lys-120 by RNF2/RING2 can also be induced by ultraviolet and may be involved in DNA repair. Following DNA double-strand breaks (DSBs), it is ubiquitinated through 'Lys-63' linkage of ubiquitin moieties by the E2 ligase UBE2N and the E3 ligases RNF8 and RNF168, leading to the recruitment of repair proteins to sites of DNA damage. Monoubiquitination and ionizing radiation-induced 'Lys-63'-linked ubiquitination are distinct events.

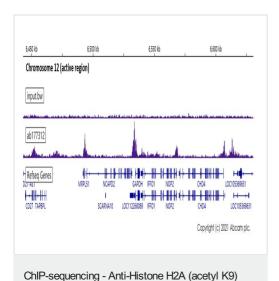
Phosphorylation on Ser-2 is enhanced during mitosis. Phosphorylation on Ser-2 by RPS6KA5/MSK1 directly represses transcription. Acetylation of H3 inhibits Ser-2 phosphorylation by RPS6KA5/MSK1.

Symmetric dimethylation on Arg-4 by the PRDM1/PRMT5 complex may play a crucial role in the germ-cell lineage.

細胞内局在

Nucleus. Chromosome.

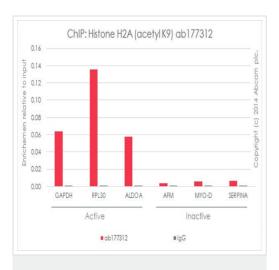
画像



antibody [EPR17471] - ChIP Grade (ab177312)

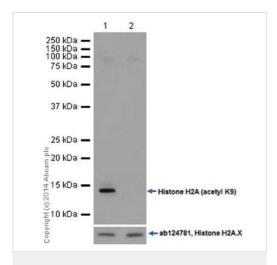
Chromatin was prepared from HeLa cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 HeLa cells and 4 µg of ab177312 (EPR17471). ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads.

Additional screenshots of mapped reads can be downloaded <u>here</u>.



ChIP - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

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 10 minutes. The ChIP was performed with 25µg of chromatin, 2µg of ab177312 (red), and 20µl of Anti rabbit IgG sepharose beads. 2µg of rabbit normal IgG was added to the beads control (grey). The immunoprecipitated DNA was quantified by real time PCR (Sybr green approach). Primers and probes are located in the first kb of the transcribed region.



Western blot - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312) **All lanes :** Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312) at 1/1000 dilution

Lane 1 : HeLa (Human epithelial cells from cervix adenocarcinoma) treated with 500 ng/ml Trichostatin A for 4 hours whole cell lysates

Lane 2 : Untreated HeLa (Human epithelial cells from cervix adenocarcinoma) whole cell lysates

Lysates/proteins at 10 µg per lane.

Secondary

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

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

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

APPL MONTREATED

APPL MELGED

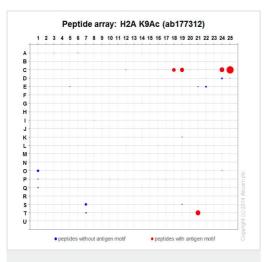
WELGED

WELGED

Immunocytochemistry/ Immunofluorescence - Anti-Histone H2A (acetyl K9) antibody [EPR17471] -ChIP Grade (ab177312) Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cells from cervix adenocarcinoma) cells, untreated and treated with Trichostatin A (500 ng/ml) for 4 hours, labeling Histone H2A (acetyl K9) with ab177312 at 1/2000 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/500 dilution (green). Confocal image showing nuclear staining on HeLa cell line. Acetylation level increased after treatment with Trichostatin A (500 ng/ml) for 4 hours. The nuclear counter stain is DAPI (blue). Tubulin is detected with ab7291 (anti-Tubulin mouse mAb) at 1/1000 dilution and ab150120 (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution (red).

The negative controls are as follows:

-ve control 1: ab177312 at 1/2000 dilution followed by <u>ab150120</u> (AlexaFluor®594 Goat anti-Mouse secondary) at 1/500 dilution. -ve control 2: <u>ab7291</u> (anti-Tubulin mouse mAb) at 1/1000 dilution followed by <u>ab150077</u> (Alexa Fluor®488 Goat Anti-Rabbit lgG H&L) at 1/500 dilution.



Peptide Array - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

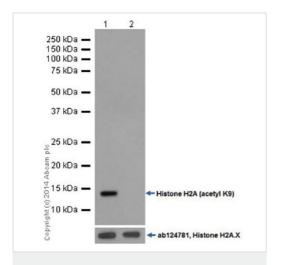
ab177312 was tested in Peptide Array against 501 different modified and unmodified histone peptides; each peptide is printed on the array at six concentrations (each in triplicate).

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

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**.



Western blot - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

All lanes : Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312) at 1/5000 dilution

Lane 1 : NIH/3T3 (Mouse embyro fibroblast cells) treated with 500 ng/ml Trichostatin A for 4 hours whole cell lysates

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

Lysates/proteins at 10 µg per lane.

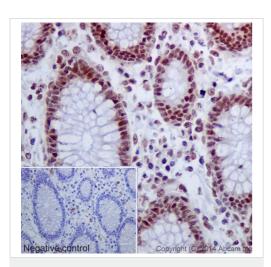
Secondary

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

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

Exposure time: 30 seconds

Blocking/Dilution buffer: 5% NFDM/TBST.

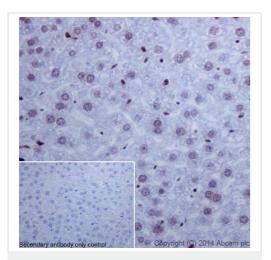


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

Immunohistochemical analysis of paraffin-embedded Human colon tissue labeling Histone H2A (acetyl K9) with ab177312 at 1/4000 dilution, followed by Goat Anti-Rabbit lgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nucleus staining on Human colon tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

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

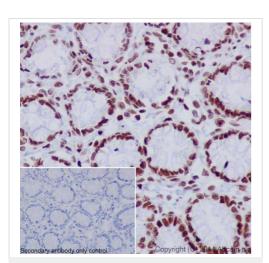


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

Immunohistochemical analysis of paraffin-embedded mouse liver tissue labeling Histone H2A (acetyl K9) with ab177312 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nucleus staining on mouse liver tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

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 H2A (acetyl K9) antibody [EPR17471] - ChIP Grade (ab177312)

Immunohistochemical analysis of paraffin-embedded rat colon tissue labeling Histone H2A (acetyl K9) with ab177312 at 1/4000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) secondary antibody at 1/500 dilution. Nucleus staining on rat colon tissue is observed. Counter stained with Hematoxylin.

Negative control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/500 dilution.

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



ChIP Grade (ab177312)

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