abcam

Product datasheet

Anti-mH2A1 antibody [EPR9359(2)] ab183041



ייבעדיו RabMAb

★★★★★ 2 Abreviews 13 References 画像数 10

製品の概要

製品名 Anti-mH2A1 antibody [EPR9359(2)]

製品の詳細 Rabbit monoclonal [EPR9359(2)] to mH2A1

由来種 Rabbit

アプリケーション 適用あり: WB, IHC-P, ICC/IF

種交差性 交差種: Mouse, Human

交差が予測される動物種: Rat 🔷

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: HepG2, MCF7, 293T and HeLa whole cell lysate (ab150035) IHC-P: Human kidney and liver

tissues ICC-IF: HAP1-WT and H2AFY knockout cells. MCF7 and 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 pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: 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

クローン名 EPR9359(2)

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ab183041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
WB		1/10000 - 1/50000. Detects a band of approximately 40 kDa (predicted molecular weight: 40 kDa).
IHC-P	****(1)	1/50 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF	*****(1)	Use a concentration of 1 µg/ml. This antibody gives positive signal in both 4%PFA and 100% MeOH-fixed cells.

ターゲット情報

Variant histone H2A which replaces conventional H2A in a subset of nucleosomes where it represses transcription. 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. Involved in stable X chromosome inactivation. Inhibits the binding of transcription factors and interferes with the activity of remodeling SWI/SNF complexes. Inhibits histone acetylation by EP300 and recruits class I HDACs, which induces an hypoacetylated state of chromatin. In addition, isoform 1, but not isoform 2, binds ADP-ribose and O-acetyl-ADP-ribose, and may be involved in ADP-ribose-mediated chromatin modulation.

組織特異性

Ubiquitous.

配列類似性

Contains 1 histone H2A domain. Contains 1 Macro domain.

翻訳後修飾

Monoubiquitinated at either Lys-116 or Lys-117. May also be polyubiquitinated. Ubiquitination is mediated by the CUL3/SPOP E3 complex and does not promote proteasomal degradation.

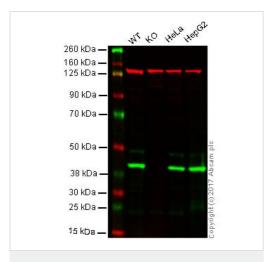
Instead, it is required for enrichment in inactive X chromosome chromatin.

細胞内局在

Nucleus. Chromosome. Enriched in inactive X chromosome chromatin and in senescence-

associated heterochromatin.

画像



Western blot - Anti-mH2A1 antibody [EPR9359(2)] (ab183041)

Lane 1: Wild type HAP1 whole cell lysate (20 µg)

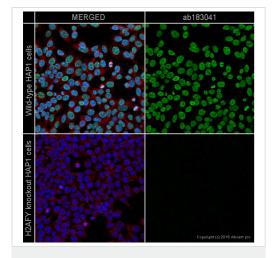
Lane 2: mH2A1 knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: Hepg2 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab183041 observed at 40 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

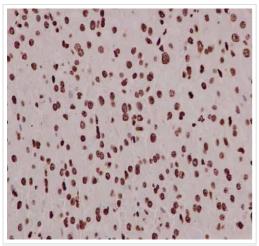
ab183041 was shown to specifically react with mH2A1 when mH2A1 knockout samples were used. Wild-type and mH2A1 knockout samples were subjected to SDS-PAGE. Ab183041 and ab18058 (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 10000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - AntimH2A1 antibody [EPR9359(2)] (ab183041)

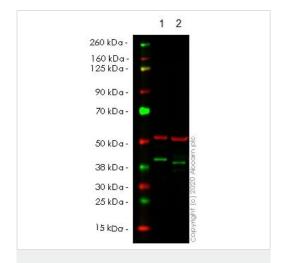
ab183041 staining mH2A1 in HAP1 WT and H2AFY knockout cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1%PBS-Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1%PBS-Tween for 1h. The cells were then incubated overnight at +4°C with ab183041 at 1µg/ml and ab195889, Mouse monoclonal [DM1A] to alpha Tubulin - Microtubule Marker (Alexa Fluor® 594) at 1/250 dilution (shown in pseudocolor red). Cells were then incubated with ab150081, Goat polyclonal Secondary Antibody to Rabbit IgG - H&L (Alexa Fluor® 488) at 1/1000 dilution (shown in green). Nuclear DNA was labelled with DAPI (shown in blue).

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-mH2A1 antibody
[EPR9359(2)] (ab183041)

Immunohistochemical analysis of paraffin-embedded Human liver tissue labeling mH2A1 with ab183041 at 1/100 dilution followed by prediluted HRP Polymer for Rabbit lgG. Counter stained with Hematoxylin.



Western blot - Anti-mH2A1 antibody [EPR9359(2)] (ab183041)

All lanes : Anti-mH2A1 antibody [EPR9359(2)] (ab183041) at 1/1000 dilution

Lane 1: Wild-type HEK-293T cell lysate

Lane 2: H2AFY CRISPR/Cas9 edited HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 40 kDa Observed band size: 40 kDa

Lanes 1-2: Merged signal (red and green). Green - ab183041 observed at 40 kDa. Red - Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) observed at 50 kDa.

ab183041 was shown to react with mH2A1 in wild-type HEK-293T cells in western blot. The band observed in CRISPR/Cas9 edited cell line ab266241 (CRISPR/Cas9 edited cell lysate ab257463) lane below 40kDa may represent truncated forms and

cleaved fragments. This has not been investigated further. Wild-type HEK-293T and H2AFY CRISPR/Cas9 edited HEK-293T cell lysates were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. ab183041 and Anti-alpha Tubulin antibody [DM1A] - Loading Control (ab7291) were incubated overnight at 4°C at a 1 in 1000 Dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse IgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

KDa 1 2 3 4
250 —
150 —
100 —
75 —
50 —
37 —
25 —
20 —
15 —
10 —

Western blot - Anti-mH2A1 antibody [EPR9359(2)] (ab183041)

All lanes : Anti-mH2A1 antibody [EPR9359(2)] (ab183041) at 1/50000 dilution

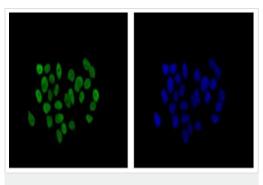
Lane 1 : HepG2 cell lysate
Lane 2 : MCF7 cell lysate
Lane 3 : HeLa cell lysate
Lane 4 : 293T cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

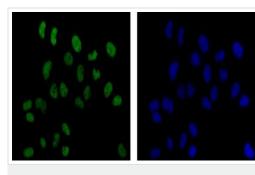
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugate at 1/1000 dilution

Predicted band size: 40 kDa **Observed band size:** 40 kDa



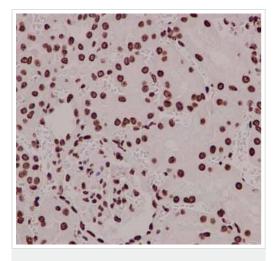
Immunocytochemistry/ Immunofluorescence - AntimH2A1 antibody [EPR9359(2)] (ab183041)

Immunofluorescent analysis of 4% paraformaldehyde-fixed MCF7 cells labeling mH2A1 with ab183041 at 1/250 dilution followed by Goat anti rabbit lgG (Alexa Fluor® 488) at 1/200 dilution (green). Counter stained with Dapi (blue).



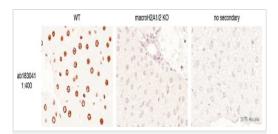
Immunocytochemistry/ Immunofluorescence - AntimH2A1 antibody [EPR9359(2)] (ab183041)

Immunofluorescent analysis of acetone-fixed HeLa cells labeling mH2A1 with ab183041 at 1/250 dilution followed by Goat anti rabbit lgG (Alexa Fluor® 488) at 1/200 dilution (green). Counter stained with Dapi (blue).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-mH2A1 antibody
[EPR9359(2)] (ab183041)

Immunohistochemical analysis of paraffin-embedded Human kidney tissue labeling mH2A1 with ab183041 at 1/100 dilution followed by prediluted HRP Polymer for Rabbit lgG. Counter stained with Hematoxylin.

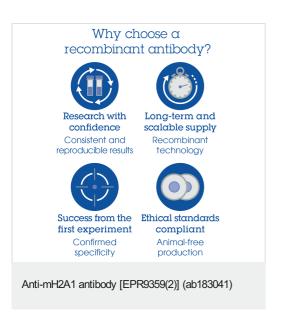


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-mH2A1 antibody

[EPR9359(2)] (ab183041)

This image is courtesy of an anonymous abreview.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of mouse liver from wild-type and mH2A1/2 knock out tissue sections labeling mH2A1 with ab183041 at 1/400 dilution. Sections were fixed in formaldehyde; heat mediated antigen retivial was performed using a citrate buffer pH 6. An undiluted polyclonal horse anti-rabbit lgG (HRP-conjugated) was used as the secondary antibody.



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