abcam

Product datasheet

Anti-Histone H2A antibody - ChIP Grade ab88770

★★★★★ 2 Abreviews 10 References 8 图像

概述

产品名称 Anti-Histone H2A抗体- ChIP Grade

描述 兔多克隆抗体to Histone H2A - ChIP Grade

宿主 Rabbit

经测试应用 适用于: ChIP, IHC-P, ICC/IF, WB, IP

种属反应性 与反应: Mouse, Cow, Human, Caenorhabditis elegans

预测可用于: Rat, Chicken, Monkey, Plasmodium falciparum 4

免疫原 Synthetic peptide corresponding to Human Histone H2A aa 50 to the C-terminus conjugated to

keyhole limpet haemocyanin. (Peptide available as <u>ab100824</u>)

阳性对照 This antibody gave a positive signal in the following whole cell lysates: Calf thymus histone; C

elegans; HeLa; NIH3T3. IHC-P: FFPE human breast adenocarcinoma tissue sections and human

colon.

常规说明

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

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C or -

80°C. Avoid freeze / thaw cycle.

存储溶液 pH: 7.40

Preservative: 0.02% Sodium azide

Constituent: PBS

Batches of this product that have a concentration < 1mg/ml may have BSA added as a stabilising

agent. If you would like information about the formulation of a specific lot, please contact our

scientific support team who will be happy to help.

1

纯度 Immunogen affinity purified

lqG

多克隆 克隆

应用

同种型

Abpromise™承诺保证使用ab88770于以下的经测试应用 The Abpromise guarantee

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
ChIP		Use 5 µg for 25 µg of chromatin.
IHC-P		Use a concentration of 0.1 - 1 µg/ml. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.
ICC/IF		Use a concentration of 1 µg/ml.
WB	*** (2)	Use a concentration of 1 µg/ml. Detects a band of approximately 17 kDa (predicted molecular weight: 14 kDa).
IP		Use a concentration of 5 µg/ml.

靶标

功能

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.

序列相似性 Belongs to the histone H2A family.

翻译后修饰 The chromatin-associated form is phosphorylated on Thr-121 during mitosis.

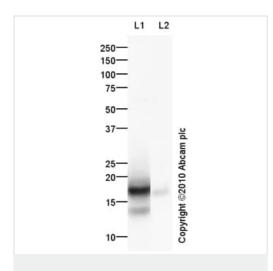
Deiminated on Arg-4 in granulocytes upon calcium entry.

Monoubiguitination 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 radiationinduced '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

图片



Western blot - Anti-Histone H2A antibody - ChIP Grade (ab88770)

All lanes : Anti-Histone H2A antibody - ChIP Grade (ab88770) at 1 μg/ml

Lane 1: Calf Thymus Histone Preparation Nuclear Lysate

Lane 2: C.elegans Tissue lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (**ab97080**) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 14 kDa **Observed band size:** 17 kDa

Additional bands at: 14 kDa. We are unsure as to the identity of

these extra bands.

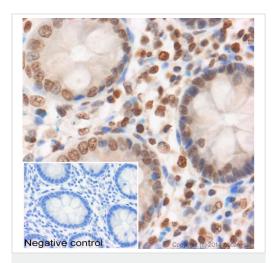
Exposure time: 2 minutes

ChIP: Histone H2A ab88770

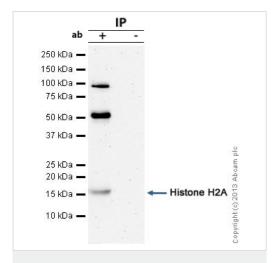
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ChIP - Anti-Histone H2A antibody - ChIP Grade (ab88770)

Chromatin was prepared from HeLa cells according to the Abcam X-ChIP protocol. Cells were fixed with formaldehyde for 10 minutes. The ChIP was performed with 25µg of chromatin, 5µg of ab88770 (blue), and 20µl of Protein A/G sepharose beads. No antibody was added to the beads control (yellow). The immunoprecipitated DNA was quantified by real time PCR (Taqman approach for active and inactive loci, Sybr green approach for heterochromatic loci). Primers and probes are located in the first kb of the transcribed region.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2A antibody - ChIP Grade (ab88770)



Immunoprecipitation - Anti-Histone H2A antibody - ChIP Grade (ab88770)

IHC image of ab88770 staining Histone H2A in human colon formalin fixed paraffin embedded tissue sections, performed on a Leica Bond. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab88770, 0.1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX. No primary antibody was used in the negative control (shown on the inset).

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.

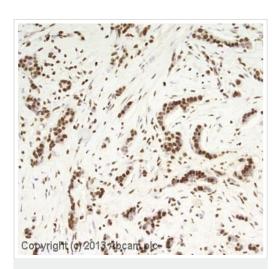
Histone H2A was immunoprecipitated using 0.5mg HeLa whole cell extract, 5µg of Rabbit polyclonal to and 50µl of protein G magnetic beads (+). No antibody was added to the control (-).

The antibody was incubated under agitation with Protein G beads for 10min, HeLa whole cell extract lysate diluted in RIPA buffer was added to each sample and incubated for a further 10min under agitation.

Proteins were eluted by addition of 40µl SDS loading buffer and incubated for 10min at 70°C; 10µl of each sample was separated on a SDS PAGE gel, transferred to a nitrocellulose membrane, blocked with 5% BSA and probed with ab88770.

Secondary: Mouse monoclonal [SB62a] Secondary Antibody to Rabbit IgG light chain (HRP) (ab99697).

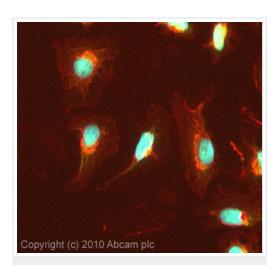
Band: 14kDa, non specific band - 95kDa: We are unsure as to the identity of this extra band; Histone H2A



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H2A antibody - ChIP Grade (ab88770)

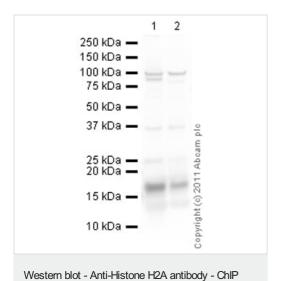
IHC image of Histone H2A staining in human breast adenocarcinoma formalin fixed paraffin embedded tissue section, performed on a Leica Bond system using the standard protocol F. The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH6, epitope retrieval solution 1) for 20 mins. The section was then incubated with ab88770, 1µg/ml, for 15 mins at room temperature and detected using an HRP conjugated compact polymer system. DAB was used as the chromogen. The section was then counterstained with haematoxylin and mounted with DPX.

For other IHC staining systems (automated and non-automated) customers should optimize variable parameters such as antigen retrieval conditions, primary antibody concentration and antibody incubation times.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H2A antibody - ChIP Grade (ab88770)

ICC/IF image of ab88770 stained HeLa cells. The cells were 100% methanol fixed (5 min) and then incubated in 1%BSA / 10% normal goat serum / 0.3M glycine in 0.1% PBS-Tween for 1h to permeabilise the cells and block non-specific protein-protein interactions. The cells were then incubated with the antibody ab88770 at 1 μ g/ml overnight at +4°C. The secondary antibody (green) was Alexa Fluor® 488 goat anti- rabbit lgG (H+L) used at a 1/1000 dilution for 1h. Alexa Fluor® 594 WGA was used to label plasma membranes (red) at a 1/200 dilution for 1h. DAPI was used to stain the cell nuclei (blue) at a concentration of 1.43 μ M. This antibody also gave a positive result in formaldehyde fixed (4%) (10min) HeLa, Hek293, HepG2, and MCF-7 cells, and in Hek293 and HepG2 Methanol (100%) (5min) fixed cells.



Grade (ab88770)

All lanes : Anti-Histone H2A antibody - ChIP Grade (ab88770) at 1 $\mu g/ml$

Lane 1 : HeLa (Human epithelial carcinoma cell line) Whole Cell Lysate

Lane 2: NIH 3T3 (Mouse embryonic fibroblast cell line) Whole Cell Lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) preadsorbed (ab97080) at 1/5000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 14 kDa **Observed band size:** 17 kDa

Additional bands at: 100 kDa, 36 kDa. We are unsure as to the

identity of these extra bands.

Exposure time: 30 seconds

Anti-Histone H2A antibody - ChIP Grade (ab88770) at 1/1000 dilution + Plasmodium falciparum histone extract at 30000000 cells

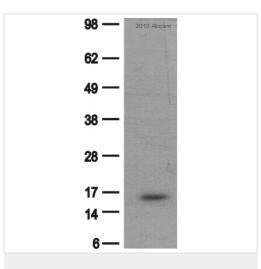
Secondary

HRP-conjugated goat anti-rabbit polyclonal IgG at 1/100000 dilution

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 14 kDa **Observed band size:** 16 kDa



Western blot - Anti-Histone H2A antibody - ChIP Grade (ab88770)

This image is courtesy of an anonymous Abreview

Exposure time: 1 minute

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