# abcam

### Product datasheet

## Anti-Histone H4 (acetyl K16) antibody [EPR1004] ab109463

重组 RabMAb

#### ★★★★★ 11 Abreviews 73 References 22 图像

#### 概述

产品名称 Anti-Histone H4 (acetyl K16)抗体[EPR1004]

描述 兔单克隆抗体[EPR1004] to Histone H4 (acetyl K16)

宿主 Rabbit

特异性 This antibody only detects Histone H4 acetylated on Lysine 16.

经测试应用 适用于: Flow Cyt (Intra), WB, IHC-P, ICC/IF, ChIC/CUT&RUN-seq

不适用于: IP

种属反应性 与反应: Mouse, Rat, Human

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

阳性对照 WB: HeLa, C6 and mouse spleen cell lysates - treated with TSA. IHC-P: Human testis, transitional

cell carcinoma and colon tissues. ICC/IF: HeLa cells treated with TSA. Flow Cyt (intra): HeLa

cells. ChIC/CUT&RUN-Seq: HeLa cells.

常规说明 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**.

#### 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.

Stable for 12 months at -20°C.

存储溶液 pH: 7.20

Preservative: 0.01% Sodium azide

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

纯度 Protein A purified

克隆 单克隆

**克隆编号** EPR1004

**同种型** IgG

#### 应用

#### The Abpromise guarantee Abpromise™承诺保证使用ab109463于以下的经测试应用

"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
Flow Cyt (Intra)		1/100 - 1/200. <b>ab172730</b> - Rabbit monoclonal lgG, is suitable for use as an isotype control with this antibody.
WB	**** (4)	1/1000 - 1/2000. Detects a band of approximately 11 kDa (predicted molecular weight: 11 kDa).
IHC-P	**** <u>(2)</u>	1/100 - 1/200. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. See IHC antigen retrieval protocols.
ICC/IF	<b>★★★★☆ (1)</b>	1/100 - 1/200.
ChlC/CUT&RUN-seq		Use at an assay dependent concentration. 2 µg

应用说明

Is unsuitable for IP.

#### 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

翻译后修饰

Belongs to the histone H4 family.

SUV420H2 and induces gene silencing.

Acetylation at Lys-6 (H4K5ac), Lys-9 (H4K8ac), Lys-13 (H4K12ac) and Lys-17 (H4K16ac) occurs in coding regions of the genome but not in heterochromatin.

Citrullination at Arq-4 (H4R3ci) by PADI4 impairs methylation.

Monomethylation and asymmetric dimethylation at Arg-4 (H4R3me1 and H4R3me2a, respectively) by PRMT1 favors acetylation at Lys-9 (H4K8ac) and Lys-13 (H4K12ac).

Demethylation is performed by JMJD6. Symmetric dimethylation on Arg-4 (H4R3me2s) by the PRDM1/PRMT5 complex may play a crucial role in the germ-cell lineage.

 $\label{lem:monomethylated} Monomethylated or trimethylated at Lys-21 (H4K20me1, H4K20me2, H4K20me3). \\ Monomethylation is performed by SET8. Trimethylation is performed by SUV420H1 and$ 

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. Monoubiquitinated at Lys-92 of histone H4 (H4K91ub1) in response to DNA damage.

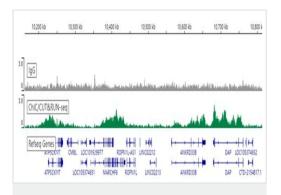
The exact role of H4K91ub1 in DNA damage response is still unclear but it may function as a

Sumoylated, which is associated with transcriptional repression.

Nucleus. Chromosome.

### 细胞定位

#### 图片

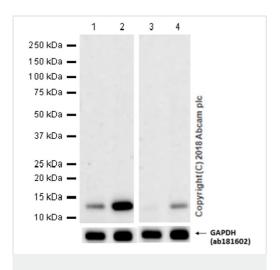


ChIC/CUT&RUN sequencing - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

ChIC/CUT&RUN was performed using a pAG-MNAse at a final concentration of 700 ng/mL,  $2.5 \times 10^5$  HeLa (Human cervix adenocarcinoma epithelial cell line) cells and  $2 \mu g$  of ab109463 [EPR1004]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control <u>ab172730</u> is also shown.

Additional screenshots of mapped reads can be downloaded **here**.

The University of Geneva owns patents relevant to ChlC (Chromatin Immuno-Cleavage) methods.



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) Lanes 1-2: Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/6000 dilution

Lanes 3-4: Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/24000 dilution

Lanes 1 & 3: Untreated C6 (Rat glial tumor glial cell) whole cell lysate

Lanes 2 & 4: C6 (Rat glial tumor glial cell) treated with Trichostatin A (final concentration is 500ng/ml) for 4 hours whole cell lysate

Lysates/proteins at 20 µg per lane.

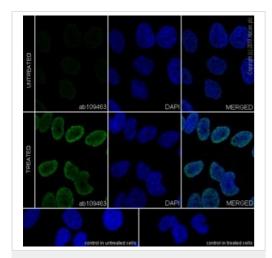
#### Secondary

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/20000 dilution

Predicted band size: 11 kDa

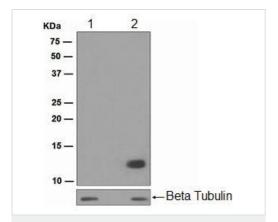
Exposure time: 3 minutes

Blocking/Diluting buffer and concentration 5% NFDM/TBST



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunocytochemistry/ Immunofluorescence analysis of untreated HeLa cells (top row) and HeLa+ TSA(500ng/ml, 4h) cells (middle row) labeling Histone H4 (acetyl K16) with ab109463 at 1/500. Goat anti rabbit lgG(Alexa Fluor<sup>®</sup> 488); **ab150077** at 1/1000 dilution was used as the secondary antibody. Cells were fixed with 4% paraformaldehyde and permeabilised with 0.1% tritonX-100. DAPI (blue) was used as a nuclear counterstain.



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

**All lanes :** Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1000 dilution (unpurified)

Lane 1: HeLa cell lysates, untreated

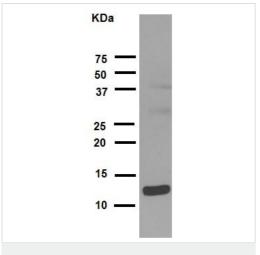
Lane 2: HeLa cell lysates treated with TSA

Lysates/proteins at 10 µg per lane.

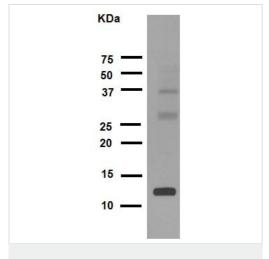
#### **Secondary**

All lanes: HRP-labelled goat anti-rabbit at 1/2000 dilution

**Predicted band size:** 11 kDa **Observed band size:** 11 kDa



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1000 dilution (unpurified) + HeLa cell lysate - treated with TSA at 10  $\mu g$ 

#### **Secondary**

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 11 kDa **Observed band size:** 11 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1500 dilution (purified) + HeLa cell lysate - treated with TSA at 10  $\mu g$ 

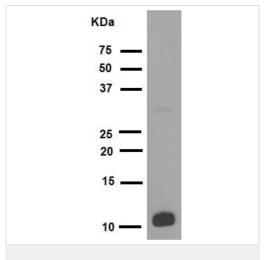
#### **Secondary**

Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

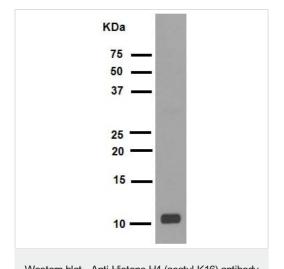
**Predicted band size:** 11 kDa **Observed band size:** 11 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1000 dilution (unpurified) + C6 cell lysate - treated with TSA at 10  $\mu g$ 

#### **Secondary**

Peroxidase-conjugated goat anti-rabbit IgG (H+L) at 1/1000 dilution

**Predicted band size:** 11 kDa **Observed band size:** 11 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1500 dilution (unpurified) + C6 cell lysate - treated with TSA at 10  $\mu g$ 

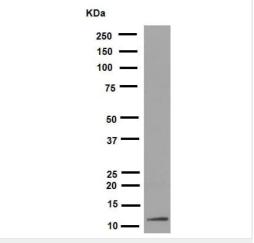
#### **Secondary**

Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

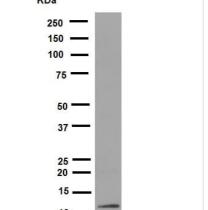
**Predicted band size:** 11 kDa **Observed band size:** 11 kDa

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)



Secondary

Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.

Peroxidase-conjugated goat anti-rabbit lgG (H+L)

Predicted band size: 11 kDa Observed band size: 11 kDa

**KDa** 250 150 100 75 ' 37 25 20 15 10

Western blot - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at 1/1500 dilution (purified) + Mouse spleen tissue lysate at 10 µg

Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463) at

1/1000 dilution (purified) + Mouse spleen tissue lysate at 10 µg

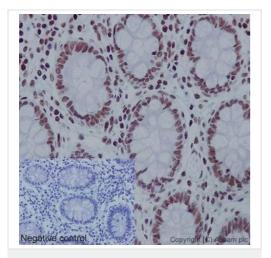
#### Secondary

Peroxidase-conjugated goat anti-rabbit lgG (H+L) at 1/1000 dilution

Predicted band size: 11 kDa Observed band size: 11 kDa

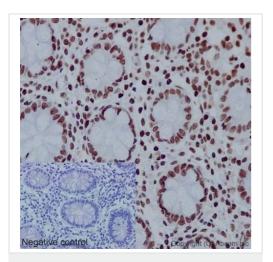
Blocking buffer and concentration: 5% NFDM/TBST.

Diluting buffer and concentration: 5% NFDM /TBST.



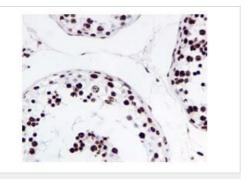
Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue labelling Histone H4 (acetyl K16) with unpurified ab109463 at 1/100. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit IgG was used as the secondary antibody. Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

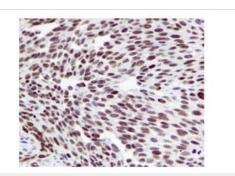
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human colon tissue labelling Histone H4 (acetyl K16) with purified ab109463 at 1/150. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. A prediluted HRP-polymer conjugated anti-rabbit lgG was used as the secondary antibody. Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human testis tissue labelling Histone H4 with unpurified ab109463 at 1/100.

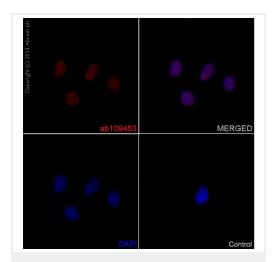
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human transitional cell carcinoma labelling Histone H4 (acetly K16) with unpurified ab109463 at 1/100.

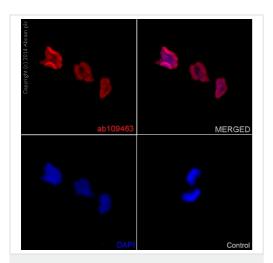
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling Histone H4 (acetyl K16) with unpurified ab109463 (red) at 1/100. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 555-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

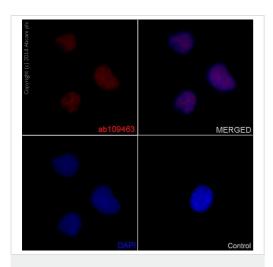
Control: primary antibody (1/100) and secondary antibody **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells treated with TSA labelling Histone H4 (acetyl K16) with unpurified ab109463 (red) at 1/100. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 555-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

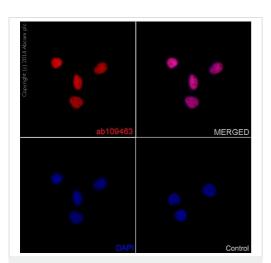
Control: primary antibody (1/100) and secondary antibody **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Immunocytochemistry/Immunofluorescence analysis of HeLa cells labelling Histone H4 (acetyl K16) with purified ab109463 (red) at 1/150. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 555-conjugated goat anti-rabbit lgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

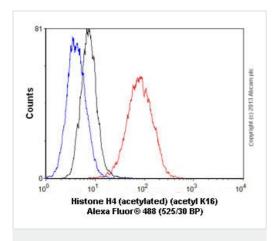
Control: primary antibody (1/150) and secondary antibody **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



Immunocytochemistry/ Immunofluorescence - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

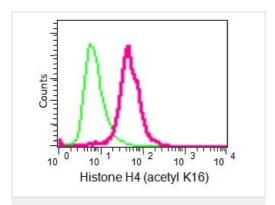
Immunocytochemistry/Immunofluorescence analysis of HeLa cells treated with TSA labelling Histone H4 (acetyl K16) with purified ab109463 (red) at 1/150. Cells were fixed with 4% paraformaldehyde. An Alexa Fluor<sup>®</sup> 555-conjugated goat anti-rabbit IgG (1/500) was used as the secondary antibody. DAPI (blue) was used as the nuclear counterstain.

Control: primary antibody (1/150) and secondary antibody **ab150120**, an Alexa Fluor<sup>®</sup> 594-conjugated goat anti-mouse IgG (1/500).



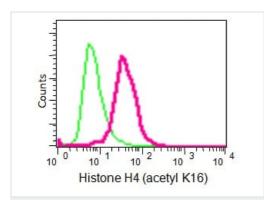
Flow Cytometry (Intracellular) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Overlay histogram showing HeLa cells stained with unpurified ab109463 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab109463, 1/1000 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluorr® 488 goat anti-rabbit IgG (H+L) (ab150077) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (0.1 $\mu$ g/1x10<sup>6</sup> cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.



Flow Cytometry (Intracellular) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Intracellular Flow Cytometry analysis of HeLa cells labelling Histone H4 (acetyl K16)with unpurified ab109463 (red) at 1/130. Cells were fixed with 80% methanol. A FITC-conjugated goat anti-rabbit lgG was used as the secondary antibody (1/150). A rabbit monoclonal lgG was used as the isotype control (green).



Flow Cytometry (Intracellular) - Anti-Histone H4 (acetyl K16) antibody [EPR1004] (ab109463)

Intracellular Flow Cytometry analysis of HeLa cells labelling Histone H4 (acetyl K16) with purified ab109463 (red) at 1/200. Cells were fixed with 80% methanol. A FITC-conjugated goat anti-rabbit lgG was used as the secondary antibody (1/150). A rabbit monoclonal lgG was used as the isotype control (green).



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