abcam

Product datasheet

Anti-HDAC2 antibody [EPR5001] - ChIP Grade ab124974





重组 RabMAb

★★★★ 4 Abreviews 10 References 8 图像

概述

产品名称 Anti-HDAC2抗体[EPR5001] - ChIP Grade

描述 兔单克隆抗体[EPR5001] to HDAC2 - ChIP Grade

宿主 Rabbit

经测试应用 适用于: WB, ICC/IF, ChIP

不适用于: Flow Cyt or IHC-P

种属反应性 与反应: Mouse, Human

预测**可用于:** Rat 4

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

阳性对照 WB: Wild-type HEK-293T, HAP1, HeLa, A431, SH-SY5Y, and Jurkat cell lysates. ICC/IF: Wild-

type HAP1 cells. ChIP: Chromatin extract from 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

存放说明 Shipped at 4°C. Store at -20°C. Stable for 12 months at -20°C.

存储溶液 pH: 7.2

Preservative: 0.05% Sodium azide

Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue

culture supernatant

纯度 Protein A purified

克隆 单克隆

克隆编号 EPR5001

同种型 IgG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab124974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
WB	★★★★ (3)	1/10000 - 1/50000. Predicted molecular weight: 55 kDa.
ICC/IF		Use a concentration of 0.5 µg/ml.
ChIP	★ की की की की (1)	Use 10 µg for 25 µg of chromatin. Please note product formulation is not optimised for ChIP application

应用说明 Is unsuitable for Flow Cyt or IHC-P.

靶标

功能 Responsible for the deacetylation of lysine residues on the N-terminal part of the core histones

(H2A, H2B, H3 and H4). Histone deacetylation gives a tag for epigenetic repression and plays an important role in transcriptional regulation, cell cycle progression and developmental events.

Histone deacetylases act via the formation of large multiprotein complexes.

Forms transcriptional repressor complexes by associating with MAD, SIN3, YY1 and N-COR. Interacts in the late S-phase of DNA-replication with DNMT1 in the other transcriptional repressor complex composed of DNMT1, DMAP1, PCNA, CAF1. Deacetylates TSHZ3 and regulates its

transcriptional repressor activity.

组织**特异性** Widely expressed; lower levels in brain and lung.

序列相似性 Belongs to the histone deacetylase family. HD type 1 subfamily.

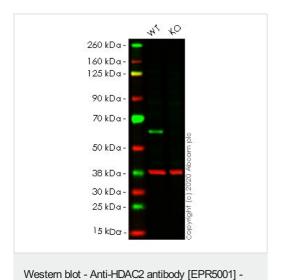
翻译后修饰 S-nitrosylated by GAPDH. In neurons, S-Nitrosylation at Cys-262 and Cys-274 does not affect the

enzyme activity but abolishes chromatin-binding, leading to increases acetylation of histones and activate genes that are associated with neuronal development. In embryonic cortical neurons, S-

Nitrosylation regulates dendritic growth and branching.

细胞定位 Nucleus.

图片



ChIP Grade (ab124974)

All lanes : Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974) at 1/10000 dilution

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

Lane 2: HDAC2 knockout HEK-293T cell lysate

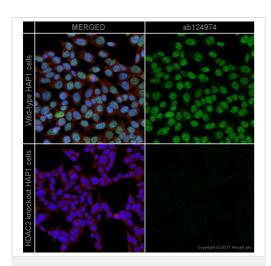
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 55 kDa **Observed band size:** 55 kDa

Lanes 1-2: Merged signal (red and green). Green - ab124974 observed at 55 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (ab8245) observed at 37 kDa.

ab124974 was shown to react with HDAC2 in wild-type HEK-293T cells in western blot. Loss of signal was observed when knockout cell line ab266589 (knockout cell lysate ab256938) was used. Wild-type HEK-293T and HDAC2 knockout 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. ab124974 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) overnight at 4°C at a 1 in 10000 dilution and a 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye®800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye®680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974)



Western blot - Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974)

ab124974 staining HDAC2 in wild-type HAP1 cells (top panel) and HDAC2 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), permeabilized with 0.1% 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 with ab124974 at 0.5μg/ml and ab195889 at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit lgG (Alexa Fluor® 488) (ab150081) at 2 μg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

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

All lanes : Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974) at 1/10000 dilution

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

Lane 2: HDAC2 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

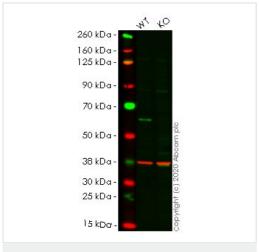
Performed under reducing conditions.

Predicted band size: 55 kDa **Observed band size:** 60 kDa

Lanes 1-2: Merged signal (red and green). Green - ab124974 observed at 60 kDa. Red - loading control **ab8245** observed at 37 kDa.

ab124974 Anti-HDAC2 antibody [EPR5001] was shown to specifically react with HDAC2 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266590 (knockout cell lysate ab2666590 (knockout cell lysate)

samples were subjected to SDS-PAGE. ab124974 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 10000 and 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.



ChIP Grade (ab124974)

All lanes : Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974) at 1/10000 dilution

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

Lane 2: HDAC2 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

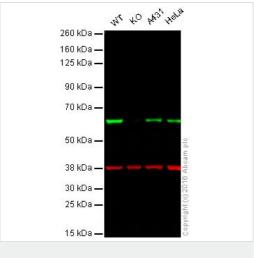
Performed under reducing conditions.

Predicted band size: 55 kDa Observed band size: 60 kDa

Western blot - Anti-HDAC2 antibody [EPR5001] -

Lanes 1-2: Merged signal (red and green). Green - ab124974 observed at 60 kDa. Red - loading control <u>ab8245</u> observed at 37 kDa.

ab124974 Anti-HDAC2 antibody [EPR5001] was shown to specifically react with HDAC2 in wild-type HEK-293T cells. Loss of signal was observed when knockout cell line ab266588 (knockout cell lysate ab256937) was used. Wild-type and HDAC2 knockout samples were subjected to SDS-PAGE. ab124974 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at 4°C at 1 in 10000 Dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit lgG H&L (IRDye® 800CW) preadsorbed (ab216773) and Goat anti-Mouse lgG H&L (IRDye® 680RD) preadsorbed (ab216776) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974)

All lanes : Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974) at 1/10000 dilution

Lane 1: Wild-type HAP1 whole cell lysate

Lane 2: HDAC2 knockout HAP1 whole cell lysate

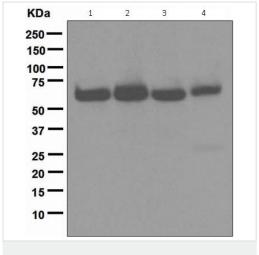
Lane 3: A431 whole cell lysate
Lane 4: HeLa whole cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 55 kDa

Lanes 1 - 4: Merged signal (red and green). Green - ab124974 observed at 60 kDa. Red - loading control, <u>ab8245</u>, observed at 37 kDa.

ab124974 was shown to recognize HDAC2 when HDAC2 knockout samples were used, along with additional cross-reactive bands. Wild-type and HDAC2 knockout samples were subjected to SDS-PAGE. ab124974 and <u>ab8245</u> (Mouse anti GAPDH loading control) were but diluted at 1/10000 and incubated overnight at 4°C. Blots were developed with 800CW Goat anti Rabbit and 680CW Goat anti Mouse secondary antibodies at 1/10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974)

All lanes : Anti-HDAC2 antibody [EPR5001] - ChIP Grade (ab124974) at 1/10000 dilution

Lane 1: HeLa cell lysate

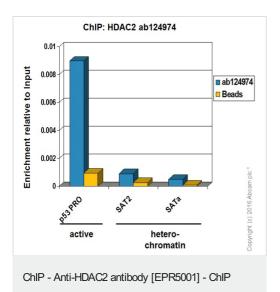
Lane 2 : SH-SY5Y cell lysate

Lane 3: Jurkat cell lysate

Lane 4: A431 cell lysate

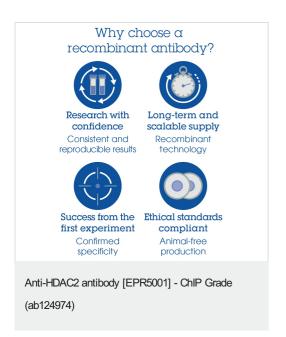
Lysates/proteins at 10 µg per lane.

Predicted band size: 55 kDa



Grade (ab124974)

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, 10µg of ab124974 (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 (Sybr green approach). Primers and probes are located in the first kb of the transcribed region.



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