abcam

Product datasheet

Anti-HDAC6 antibody [EPR1698(2)] ab133493

KO VALIDATED Recombinant RabMAb

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Overview

Product name	Anti-HDAC6 antibody [EPR1698(2)]
Description	Rabbit monoclonal [EPR1698(2)] to HDAC6
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IHC-P
Species reactivity	**Reacts with:** Human, African green monkey **Predicted to work with:** Monkey
Immunogen	Synthetic peptide within Human HDAC6 aa 1-100 (N terminal). The exact sequence is proprietary. Database link: **Q9UBN7**
Positive control	WB: HAP1, HeLa, Jurkat, and COS-1 cell lysates. Flow Cyt (intra): K562. IHC-P: Human kidney tissue.
General notes	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**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.05% Sodium azide Constituents: 0.1% BSA, 40% Glycerol (glycerin, glycerine), 9.85% Tris glycine, 50% Tissue culture supernatant
Purity	Tissue culture supernatant

Clonality	Monoclonal
Clone number	EPR1698(2)
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab133493 in the following tested applications.

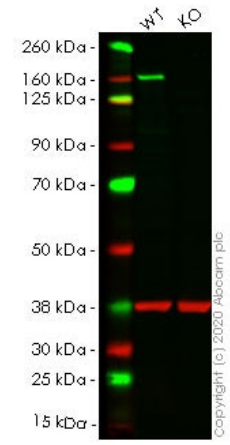
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/200.
WB	★★★☆☆ **(1)**	1/10000 - 1/50000. Detects a band of approximately 160 kDa (predicted molecular weight: 131 kDa).
IHC-P		1/50 - 1/100. Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.

Target

Function	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 (By similarity). Plays a central role in microtubule-dependent cell motility via deacetylation of tubulin.
Sequence similarities	Belongs to the histone deacetylase family. HD type 2 subfamily. Contains 1 UBP-type zinc finger.
Post-translational modifications	Ubiquitinated. Its polyubiquitination however does not lead to its degradation. Sumoylated in vitro.
Cellular localization	Nucleus. Cytoplasm. It is mainly cytoplasmic, where it is associated with microtubules.

Images



Western blot - Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

All lanes : Anti-HDAC6 antibody [EPR1698(2)] (ab133493) at 1/10000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : HDAC6 knockout HeLa cell lysate

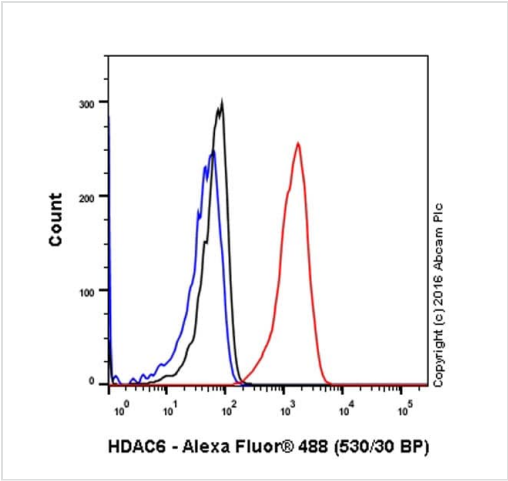
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 131 kDa
Observed band size: 160 kDa

Lanes 1- 2: Merged signal (red and green). Green - ab133493 observed at 160 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab133493 was shown to react with HDAC6 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line was used. Wild-type HeLa and HDAC6 knockout HeLa 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. ab133493 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 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.

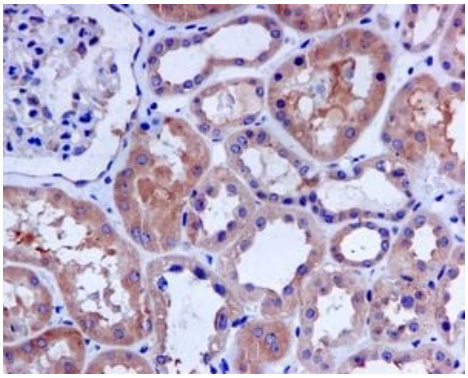


Flow Cytometry (Intracellular) - Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

ab133493 staining HDAC6 in K562 (human chronic myelogenous leukemia) by intracellular flow cytometry. Cells were fixed with 4% paraformaldehyde, permabilised with 90% methanol and the sample was incubated with the primary antibody at a dilution of 1/200. A goat anti rabbit IgG (Alexa Fluorr® 488) at a dilution of 1/2000 was used as the secondary antibody.

Isoytype control: Rabbit monoclonal IgG (Black)

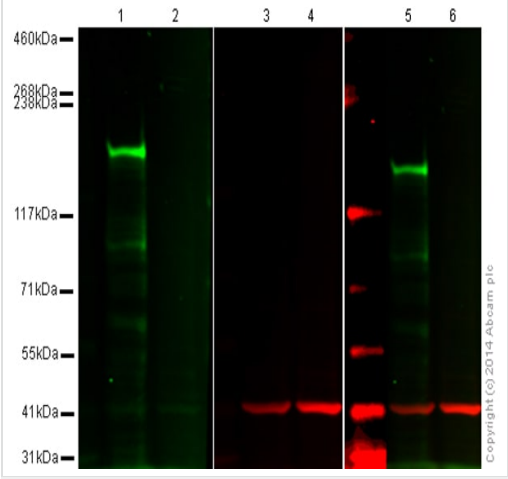
Unlabelled control: Cell without incubation with primary antibody and secondary antibody (Blue)



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

Immunohistochemical analysis of paraffin embedded Human kidney tissue labelling HDAC6 with ab133493 antibody at a dilution of 1/50.

Perform heat mediated antigen retrieval with citrate buffer pH 6 before commencing with IHC staining protocol.



Western blot - Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

Lanes 1-2 : Anti-HDAC6 antibody [EPR1698(2)] (ab133493) at 1/10000 dilution
Lanes 3-4 : Anti-beta Actin antibody [mAbcam 8226] - Loading Control (**ab8226**) at 1/1000 dilution

Lanes 1 & 3 & 5 : Wild-type HAP1 cell lysate
Lanes 2 & 4 & 6 : HDAC6 knockout HAP1 cell lysate

Lysates/proteins at 20 µg per lane.

Predicted band size: 131 kDa

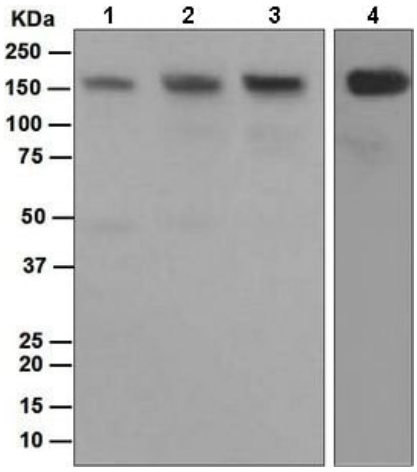
Lanes 1 and 2: Green signal from target – ab133493 observed at

160 kDa

Lanes 3 and 4: Red signal from loading control – **ab8226** observed at 42 kDa

Lanes 5 and 6: Merged (red and green) signal

ab133493 was shown to specifically react with HDAC6 when HDAC6 knockout samples were used. Wild-type and HDAC6 knockout samples were subjected to SDS-PAGE. ab133493 and **ab8226** (loading control to beta actin) were diluted 1/10 000 and 1/1000 respectively and incubated overnight at 4ºC. 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 1/10 000 dilution for 1 h at room temperature before imaging.



Western blot - Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

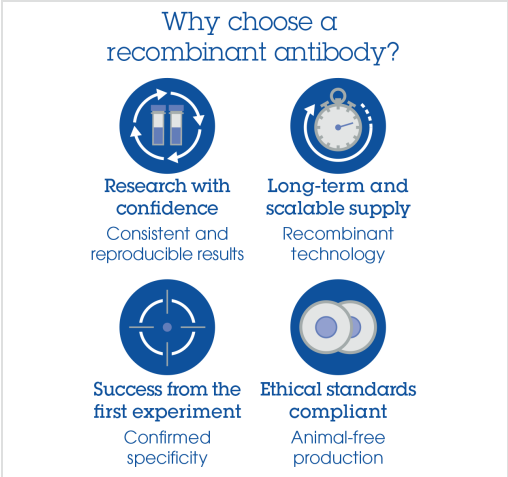
All lanes : Anti-HDAC6 antibody [EPR1698(2)] (ab133493) at 1/10000 dilution

Lane 1 : HeLa cell lysate
Lane 2 : Jurkat cell lysate
Lane 3 : K562 cell lysate
Lane 4 : COS-1 cell lysate

Lysates/proteins at 10 µg per lane.

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

Predicted band size: 131 kDa
Observed band size: 160 kDa



Anti-HDAC6 antibody [EPR1698(2)] (ab133493)

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