

Product datasheet

Anti-ALKBH5 antibody [EPR18958] ab195377

KO VALIDATED Recombinant RabMAb

★★★★★ 1 Abreviews 14 References 6 Images

Overview

Product name	Anti-ALKBH5 antibody [EPR18958]
Description	Rabbit monoclonal [EPR18958] to ALKBH5
Host species	Rabbit
Specificity	ab195377 has been tested in IHC-P on Human, Mouse and Rat testis sections. It worked well on Human, but not on the Mouse or Rat sections, under our experimental conditions.
Tested applications	Suitable for: WB, IHC-P
Species reactivity	Reacts with: Mouse, Rat, Human Predicted to work with: Sheep, Goat, Horse, Guinea pig, Cow, Cat, Dog, Pig, Non human primates
Immunogen	Recombinant fragment within Human ALKBH5 aa 50-350. The exact sequence is proprietary. Database link: Q6P6C2
Positive control	WB: Wild type MEF, HeLa, HepG2 , PC-12, NIH/3T3, Jurkat, Neuro-2a, HEK-293T and F9 whole cell lysates. IHC-P: Human testis and kidney tissues.
General notes	<p>This product is a recombinant monoclonal antibody, which offers several advantages including:</p> <ul style="list-style-type: none"> - High batch-to-batch consistency and reproducibility - Improved sensitivity and specificity - Long-term security of supply - Animal-free production <p>For more information see here.</p> <p>Our RabMAb[®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb[®] patents.</p> <p>Reproducibility is key to advancing scientific discovery and accelerating scientists' next breakthrough.</p> <p>Abcam is leading the way with our range of recombinant antibodies, knockout-validated antibodies and knockout cell lines, all of which support improved reproducibility.</p> <p>We are also planning to innovate the way in which we present recommended applications and species on our product datasheets, so that only applications & species that have been tested in our own labs, our suppliers or by selected trusted collaborators are covered by our Abpromise[™] guarantee.</p> <p>In preparation for this, we have started to update the applications & species that this product is</p>

Abpromise guaranteed for.

We are also updating the applications & species that this product has been “predicted to work with,” however this information is not covered by our Abpromise guarantee.

Applications & species from publications and Abreviews that have not been tested in our own labs or in those of our suppliers are not covered by the Abpromise guarantee.

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, as well as customer reviews and Q&As.

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 long term. Avoid freeze / thaw cycle.
Storage buffer	pH: 7.2 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18958
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab195377** in the following tested applications.

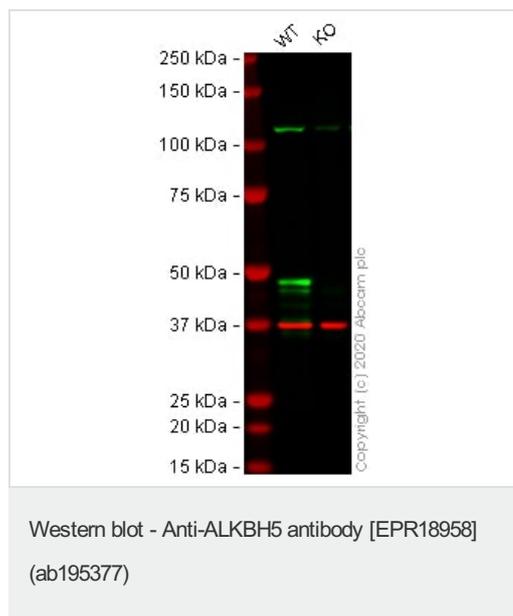
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
WB	★★★★★	1/1000. Detects a band of approximately 35-44 kDa (predicted molecular weight: 44 kDa).
IHC-P		1/2000. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol. Suitable for human tissues only.

Target

Relevance	Dioxygenase that demethylates RNA by oxidative demethylation: specifically demethylates N(6)-methyladenosine (m6A) RNA, the most prevalent internal modification of messenger RNA (mRNA) in higher eukaryotes. Requires molecular oxygen, alpha-ketoglutarate and iron. Demethylation of m6A mRNA affects mRNA processing and export and is required for spermatogenesis.
Cellular localization	Nucleus speckle

Images



All lanes : Anti-ALKBH5 antibody [EPR18958] (ab195377) at 1/1000 dilution

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

Lane 2 : ALKBH5 knockout HEK-293T cell lysate

Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

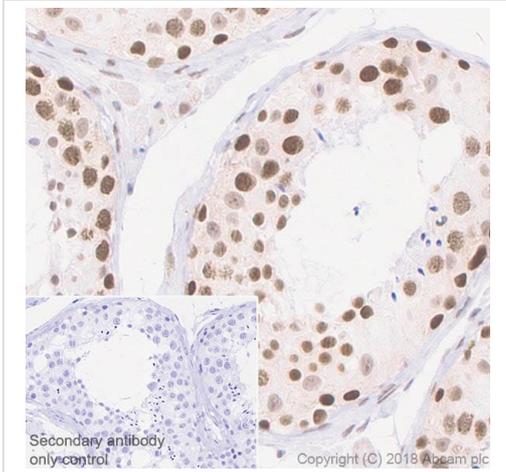
Predicted band size: 44 kDa

Observed band size: 48 kDa

[why is the actual band size different from the predicted?](#)

Lanes 1 - 2: Merged signal (red and green). Green - ab195377 observed at 48 kDa. Red - loading control [ab8245](#) (Mouse anti-GAPDH antibody [6C5]) observed at 37kDa.

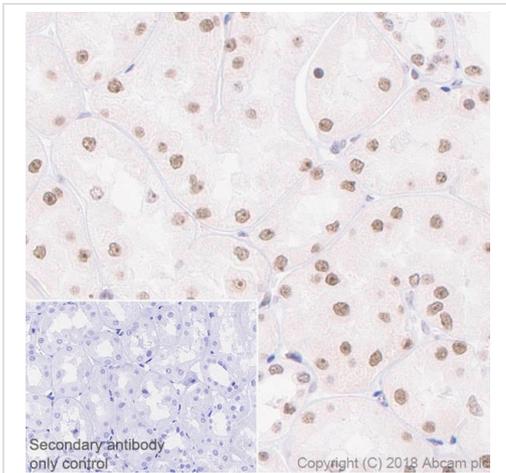
ab195377 was shown to react with ALKBH5 in wild-type HEK-293T cells in western blot with loss of signal observed in ALKBH5 knockout cell line [ab266762](#) (ALKBH5 knockout cell lysate [ab257349](#)). Wild-type and ALKBH5 knockout HEK-293T cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3% milk in TBS-T (0.1% Tween[®]) before incubation with ab195377 and [ab8245](#) (Mouse anti-GAPDH antibody [6C5]) overnight at 4°C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Mouse IgG H&L (IRDye[®] 800CW) preabsorbed ([ab216772](#)) and Goat anti-Rabbit IgG H&L (IRDye[®] 680RD) preabsorbed ([ab216777](#)) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ALKBH5 antibody [EPR18958] (ab195377)

Immunohistochemical analysis of paraffin-embedded human testis tissue labeling ALKBH5 with ab195377 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear staining in human testis (PMID: 21264265; PMID: 27590511; PMID: 17979886) is observed. Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

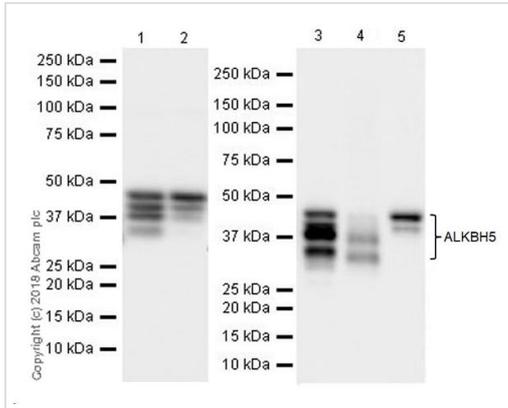
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-ALKBH5 antibody [EPR18958] (ab195377)

Immunohistochemical analysis of paraffin-embedded human kidney tissue labeling ALKBH5 with ab195377 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use. Nuclear staining in human kidney (PMID: 21264265) is observed. Counter stained with hematoxylin. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.



Western blot - Anti-ALKBH5 antibody [EPR18958] (ab195377)

Lanes 1-2 : Anti-ALKBH5 antibody [EPR18958] (ab195377) at 1/1000 dilution

Lanes 3-5 : Anti-ALKBH5 antibody [EPR18958] (ab195377) at 1/2000 dilution

Lane 1 : PC-12 (rat adrenal gland pheochromocytoma cell line) whole cell lysate

Lane 2 : NIH/3T3 (mouse embryo fibroblast cell line) whole cell lysate

Lane 3 : Jurkat (human T cell leukemia cell line from peripheral blood) whole cell lysate

Lane 4 : Neuro-2a (mouse neuroblastoma cell line) whole cell lysate

Lane 5 : F9 (mouse embryonic testicular cancer cell line) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

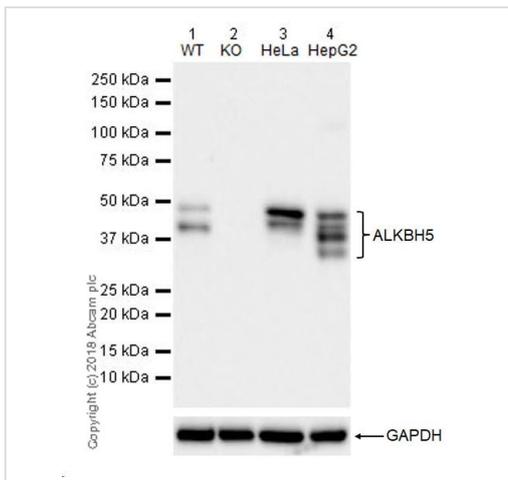
Predicted band size: 44 kDa

Observed band size: 35-44 kDa [why is the actual band size different from the predicted?](#)

Exposure time : Lanes 1 and 2: 15 seconds; Lanes 3 and 4: 26 seconds.

Blocking/Dilution buffer: 5% NFDm/TBST.

The molecular mass observed is consistent with the literature (PMID 21264265).



Western blot - Anti-ALKBH5 antibody [EPR18958] (ab195377)

All lanes : Anti-ALKBH5 antibody [EPR18958] (ab195377) at 1/5000 dilution

Lane 1 : Wild type MEF whole cell lysate

Lane 2 : ALKBH5 knockout MEF whole cell lysate

Lane 3 : HeLa (human epithelial cell line from cervix adenocarcinoma) whole cell lysate

Lane 4 : HepG2 (human liver hepatocellular carcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Developed using the ECL technique.

Predicted band size: 44 kDa

Observed band size: 35-44 kDa [why is the actual band size different from the predicted?](#)

Exposure time: 70 seconds

Blocking/Dilution buffer: 5% NFDm/TBST.

The Wt and ALKBH5 KO cell lysates were kindly provided by an anonymous collaborator.

The molecular mass observed is consistent with the literature (PMID 21264265).

Why choose a recombinant antibody?



Research with confidence
Consistent and reproducible results



Long-term and scalable supply
Recombinant technology



Success from the first experiment
Confirmed specificity



Ethical standards compliant
Animal-free production

Anti-ALKBH5 antibody [EPR18958] (ab195377)

Please note: All products are "FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES"

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