abcam

ab249990

Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free

Recombinant | RabMAb | Advanced Validation | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR12742
Purification technique	Affinity purification Protein A
Concentration	1.04 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Mouse
Dilution info	-

Notes	-

Not recommended

Species	Human, Rat
Dilution info	-
Notes	-

ChIC/CUT&RUN-seq

Tested

Species	Human, Mouse
Dilution info	-
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

IHC-P

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

ICC/IF

Not recommended

Species	Human, Mouse, Rat
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

Expected

Species	Mouse
Dilution info	Use at an assay dependent concentration.
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

ChIP-seq

Tested

Species	Human, Mouse
Dilution info	-
Notes	-

Predicted

Species	Rat
Dilution info	-
Notes	-

Target data

See full target information KDM5A

Function

Histone demethylase that specifically demethylates 'Lys-4' of histone H3, thereby playing a central role in histone code. Does not demethylate histone H3 'Lys-9', H3 'Lys-27', H3 'Lys-36', H3 'Lys-79' or H4 'Lys-20'. Demethylates trimethylated and dimethylated but not monomethylated H3 'Lys-4'. Regulates specific gene transcription through DNA-binding on 5'-CCGCCC-3' motif (PubMed:18270511). May stimulate transcription mediated by nuclear receptors. Involved in transcriptional regulation of Hox proteins during cell differentiation (PubMed:19430464). May participate in transcriptional repression of cytokines such as CXCL12. Plays a role in the regulation of the circadian rhythm and in maintaining the normal periodicity of the circadian clock. In a histone demethylase-independent manner, acts as a coactivator of the CLOCK-BMAL1-mediated transcriptional activation of PER1/2 and other clock-controlled genes and increases histone acetylation at PER1/2 promoters by inhibiting the activity of HDAC1 (By similarity). Seems to act as a transcriptional corepressor for some genes such as MT1F and to favor the proliferation of cancer cells (PubMed:27427228).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab249990 is the carrier-free version of ab177486.

Patented technology
Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMAb® patents.

What are the advantages of a recombinant monoclonal antibody?
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 batch production

For more information, read more on recombinant antibodies.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

We do not recommend this combination. It is not covered by our product promise.

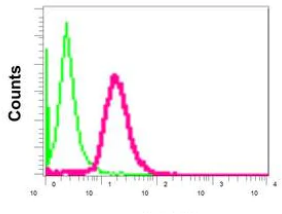
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

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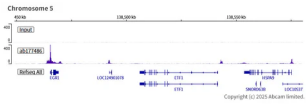
16 product images



Flow Cytometry (Intracellular) - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

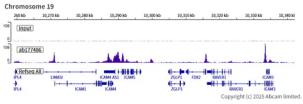
KDM5A / Jarid1A / RBBP2 Flow Cytometry (Intracellular) staining using rabbit Anti-KDM5A / Jarid1A / RBBP2 antibody

This data was developed using ab177486, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of permeabilized NCCIT cells labeling KDM5A / Jarid1A / RBBP2 with ab177486 at 1/10 dilution (red) or a rabbit IgG (negative) (green).



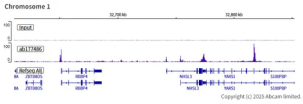
ChIP-sequencing - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

This data was developed using ab177486, the same antibody clone in a different buffer formulation.
Chromatin was prepared from K-562 cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 10^7 cells and 8 µg of ab177486 [EPR12742]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads. The Input control is also shown.



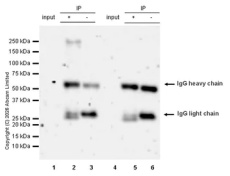
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Immunoprecipitation - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

This data was developed using ab177486, the same antibody clone in a different buffer formulation.

KDM5A / Jarid1A / RBBP2 was immunoprecipitated from 0.35 mg NIH/3T3 (mouse embryonic fibroblast) whole cell lysate with ab177486 at 1/30 dilution (2µg in 0.35mg lysates). Western blot was performed on the immunoprecipitate using ab177486 at 1/1000 dilution.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

This blot was developed using a high-sensitivity ECL substrate, allowing for the detection of proteins in the mid-femtogram range.
All lanes:
Immunoprecipitation - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] (ab177486) at 1/30 dilution

Lane 1:
3T3-L1 (mouse embryonic fibroblast) transfected with scrambled siRNA control whole cell lysate at 7 µg

Lane 2:
3T3-L1 transfected with scrambled siRNA control whole cell lysate

Lane 3:
Rabbit monoclonal IgG (ab172730) instead of ab194286 in 3T3-L1 transfected with scrambled siRNA control lysate

Lane 4:
3T3-L1 transfected with siRNA specifically targeting KDM5A whole cell lysate at 7 µg

Lane 5:
3T3-L1 transfected with siRNA specifically targeting KDM5A whole cell lysate

Lane 6:

Rabbit monoclonal IgG (ab172730) instead of ab194286 in 3T3-L1 transfected with siRNA specifically targeting KDM5A lysate

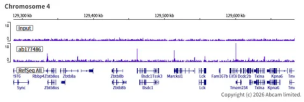
Secondary

All lanes:
Immunoprecipitation - VeriBlot for IP Detection Reagent (HRP) (ab131366) at 1/5000 dilution

Developed using the ECL technique.

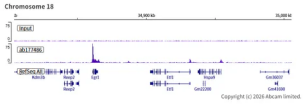
Observed band size: 200 kDa

Exposure time: 180s



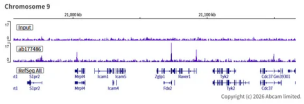
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This data was developed using ab177486, the same antibody clone in a different buffer formulation.
Chromatin was prepared from 3T3-L1 cells. Cells were fixed with 1% formaldehyde for 10 minutes. ChIP was performed with 5 x 10^6 cells and 4 µg of ab177486 [EPR12742]. ChIP DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 30 million reads. The Input control is also shown.



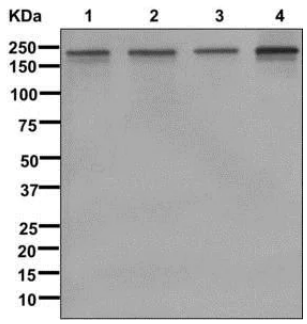
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Western blot - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

This data was developed using ab177486, the same antibody clone in a different buffer formulation.
All lanes:
Western blot - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] (ab177486) at 1/1000 dilution

Lane 1:

HeLa cell lysate at 10 µg

Lane 2:
293T cell lysate at 10 µg

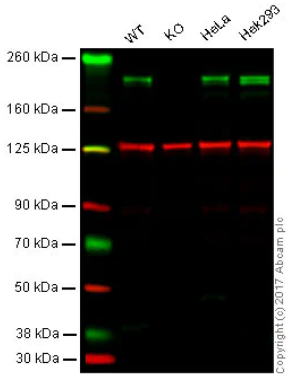
Lane 3:
MCF7 cell lysate at 10 µg

Lane 4:
NCCIT cell lysate at 10 µg

Secondary

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

Predicted band size: 192 kDa



Western blot - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

KDM5A / Jarid1A / RBBP2 Western blot staining using rabbit Anti-KDM5A / Jarid1A / RBBP2 antibody

This data was developed using ab177486, the same antibody clone in a different buffer formulation.
Lane 1: Wild-type HAP1 whole cell lysate (20 µg)

Lane 2: KDM5A knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

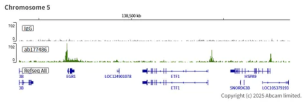
Lane 4: HEK293 whole cell lysate (20 µg)

Lanes 1 - 4: Merged signal (red and green). Green - ab177486 observed at 240 kDa. Red - loading control, ab18058, observed at 130 kDa.

ab177486 was shown to specifically react with KDM5A in wild-type HAP1 cells. No bands were observed when KDM5A knockout samples were used. Wild-type and KDM5A knockout samples were subjected to SDS-PAGE. ab177486 and ab18058 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/10000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed ab216776 secondary antibodies at 1/20000 dilution for 1 hour at room temperature before imaging.

All lanes:
Western blot - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] (ab177486)

Predicted band size: 192 kDa

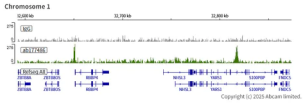


ChIC/CUT&RUN sequencing - Anti-KDM5A / Jarid1A / RBBP2 antibody [EPR12742] - BSA and Azide free (ab249990)

This data was developed using ab177486, the same antibody clone in a different buffer formulation.

ChIC/CUT&RUN was performed using a pAG-MNase at a final concentration of 700 ng/mL, 2.5 x 10^5 K-562 (human chronic myelogenous leukemia lymphoblast) cells and 5 µg of ab177486

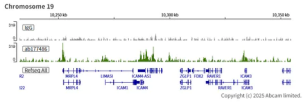
[EPR12742]. The resulting DNA was sequenced on the Illumina NovaSeq 6000 to a depth of 10 million reads. The negative IgG control ab172730 is also shown. The University of Geneva owns patents relevant to ChIC (Chromatin Immuno-Cleavage) methods.



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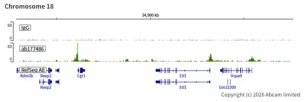
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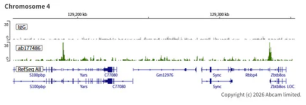
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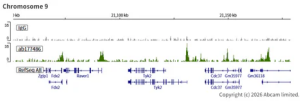
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Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.