abcam

Product datasheet

Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free ab251059

KO VALIDATED Recombinant RabMAb

8 Images

Overview

Product name	Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free
Description	Rabbit monoclonal [EPR18875] to KDM3A / JHDM2A - BSA and Azide free
Host species	Rabbit
Tested applications	**Suitable for:** IP, WB, IHC-P
Species reactivity	**Reacts with:** Human
Immunogen	Synthetic peptide. This information is proprietary to Abcam and/or its suppliers.
General notes	ab251059 is the carrier-free version of **ab191389**. Our **carrier-free** antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. The carrier-free buffer and high concentration allow for increased conjugation efficiency. 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. 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. 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

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C. Do Not Freeze.
Storage buffer	pH: 7.2 Constituent: PBS
Carrier free	Yes
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18875
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab251059 in the following tested applications.

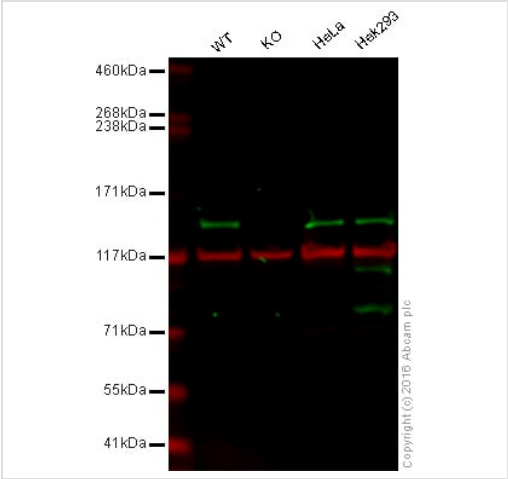
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
IP		Use at an assay dependent concentration.
WB		Use at an assay dependent concentration. Detects a band of approximately 147, 100, 75 kDa (predicted molecular weight: 147 kDa).
IHC-P		Use at an assay dependent concentration. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.

Target

Function	Histone demethylase that specifically demethylates 'Lys-9' of histone H3, thereby playing a central role in histone code. Preferentially demethylates mono- and dimethylated H3 'Lys-9' residue, with a preference for dimethylated residue, while it has weak or no activity on trimethylated H3 'Lys-9'. Demethylation of Lys residue generates formaldehyde and succinate. Involved in hormone-dependent transcriptional activation, by participating in recruitment to androgen-receptor target genes, resulting in H3 'Lys-9' demethylation and transcriptional activation. Involved in spermatogenesis by regulating expression of target genes such as PRM1 and TMP1 which are required for packaging and condensation of sperm chromatin. Involved in obesity resistance through regulation of metabolic genes such as PPARA and UCP1.
Sequence similarities	Belongs to the JHDM2 histone demethylase family. Contains 1 JmjC domain.
Domain	The JmjC domain and the C6-type zinc-finger are required for the demethylation activity. Leu-Xaa-Xaa-Leu-Leu (LXXLL) motifs are known to mediate the association with nuclear receptors.
Cellular localization	Cytoplasm. Nucleus. Nuclear in round spermatids. When spermatids start to elongate, localizes to the cytoplasm where it forms distinct foci which disappear in mature spermatozoa.

Images



Western blot - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.

Lane 1: Wild type HAP1 whole cell lysate (40 µg)

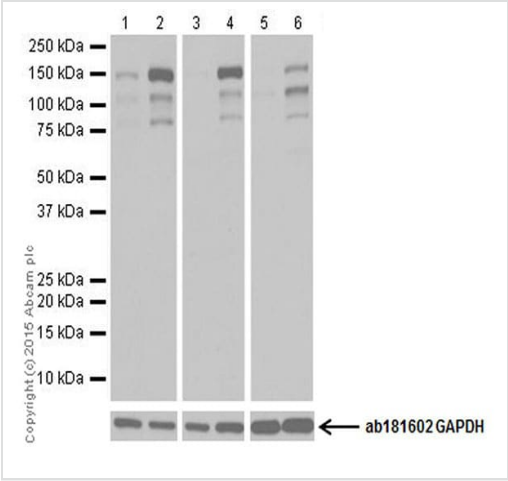
Lane 2: KDM3A knockout HAP1 whole cell lysate (40 µg)

Lane 3: HeLa whole cell lysate (40 µg)

Lane 4: Hek293 whole cell lysate (40 µg)

Lanes 1 - 4: Merged signal (red and green). Green - **ab191389** observed at 150 kDa. Red - loading control, **ab18058**, observed at 130 kDa.

ab191389 was shown to specifically react with KDM3A when KDM3A knockout samples were used. Wild-type and KDM3A knockout samples were subjected to SDS-PAGE. **ab191389** and **ab18058** (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 500 dilution and 1/10000 dilution respectively. 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-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

All lanes : Anti-KDM3A / JHDM2A antibody [EPR18875] (**ab191389**) at 1/1000 dilution

Lane 1 : Untreated HEK-293 (Human epithelial cell line from embryonic kidney), whole cell lysate
Lane 2 : HEK-293 (Human epithelial cell line from embryonic kidney) treated with 100 µM CoCl2 for 24 hours, whole cell lysate
Lane 3 : Untreated HeLa (Human epithelial cell line from cervix adenocarcinoma), whole cell lysate
Lane 4 : HeLa (Human epithelial cell line from cervix adenocarcinoma) treated with 100 µM CoCl2 for 24 hours, whole cell lysate
Lane 5 : Untreated Jurkat (Human T cell leukemia cell line from peripheral blood), whole cell lysate
Lane 6 : Jurkat (Human T cell leukemia cell line from peripheral blood) treated with 100 µM CoCl2 for 24 hours, whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at

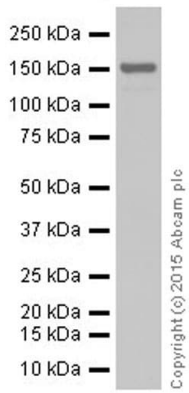
1/100000 dilution

Predicted band size: 147 kDa
Observed band size: 100,147,75 kDa

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.

Exposure times: Lanes 1 and 2: 15 seconds; Lanes 3 and 4: 30 seconds; Lanes 5 and 6: 5 seconds.



Western blot - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

Anti-KDM3A / JHDM2A antibody [EPR18875] (**ab191389**) at 1/1000 dilution + Ramos (Human Burkitt's lymphoma cell line) whole cell lysate

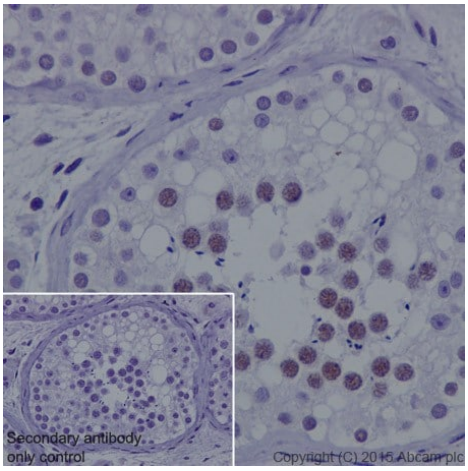
Secondary
Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/100000 dilution

Predicted band size: 147 kDa
Observed band size: 147 kDa

Exposure time: 3 minutes

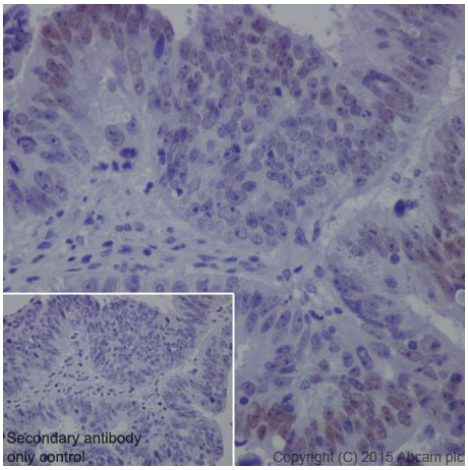
This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.

Blocking and dilution buffer: 5% NFDM/TBST.



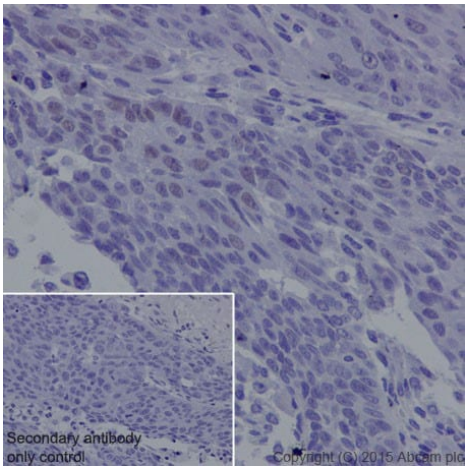
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.Immunohistochemical analysis of paraffin-embedded human testis tissue labeling KDM3A / JHDM2A with **ab191389** at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Nuclear staining on spermatogenic cells of human testis is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



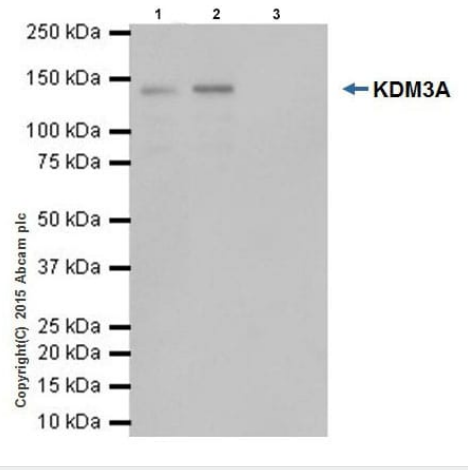
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.Immunohistochemical analysis of paraffin-embedded human bladder cancer tissue labeling KDM3A / JHDM2A with **ab191389** at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Weak nuclear staining on cancer cells of human bladder cancer is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



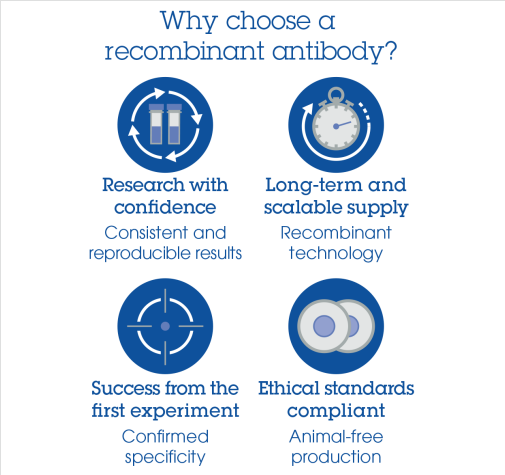
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.Immunohistochemical analysis of paraffin-embedded human lung cancer tissue labeling KDM3A / JHDM2A with **ab191389** at 1/1000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Weak nuclear staining on cancer cells of human lung cancer is observed. Counter stained with Hematoxylin. Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/500 dilution. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunoprecipitation - Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

This data was developed using **ab191389**, the same antibody clone in a different buffer formulation.KDM3A / JHDM2A was immunoprecipitated from 1mg of Ramos (Human Burkitt's lymphoma cell line) whole cell lysate with **ab191389** at 1/30 dilution. Western blot was performed from the immunoprecipitate using **ab191389** at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (**ab131366**), was used for detection at 1/10000 dilution. Lane 1: Ramos whole cell lysate 10µg (Input). Lane 2: **ab191389** IP in Ramos whole cell lysate. Lane 3: Rabbit monoclonal IgG (**ab172730**) instead of **ab191389** in Ramos whole cell lysate. Blocking and dilution buffer and concentration: 5% NFDM/TBST. Exposure time: 10 seconds.



Anti-KDM3A / JHDM2A antibody [EPR18875] - BSA and Azide free (ab251059)

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