abcam

Product datasheet

Anti-KDM6A / UTX antibody [EPR23203-211] ab253183

KO VALIDATED Recombinant RabMAb

5 Images

Overview

Product name	Anti-KDM6A / UTX antibody [EPR23203-211]
Description	Rabbit monoclonal [EPR23203-211] to KDM6A / UTX
Host species	Rabbit
Tested applications	**Suitable for:** Flow Cyt (Intra), WB **Unsuitable for:** ChIP,ICC/IF,IHC-P or IP
Species reactivity	**Reacts with:** Mouse, Human
Immunogen	Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.
Positive control	WB: HAP1, HEK-293T, NIH/3T3 and F9 whole cell lysates. Flow Cyt (intra): HAP1 and F9 cells.
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 long term. Avoid freeze / thaw cycle.
Storage buffer	Preservative: 0.01% Sodium azide Constituents: 59.94% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR23203-211
Isotype	IgG

Applications

The Abpromise guarantee Our Abpromise guarantee covers the use of ab253183 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)		1/50.
WB		1/1000. Detects a band of approximately 160 kDa (predicted molecular weight: 154 kDa).

Application notes Is unsuitable for ChIP,ICC/IF,IHC-P or IP.

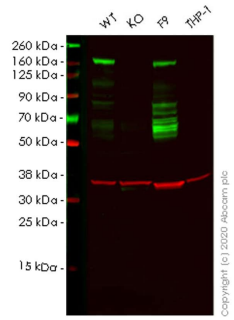
Target

Function Histone demethylase that specifically demethylates 'Lys-27' of histone H3, thereby playing a central role in histone code. Demethylates trimethylated and dimethylated but not monomethylated H3 'Lys-27'. Plays a central role in regulation of posterior development, by regulating HOX gene expression. Demethylation of 'Lys-27' of histone H3 is concomitent with methylation of 'Lys-4' of histone H3, and regulates the recruitment of the PRC1 complex and monoubiquitination of histone H2A.

Sequence similarities Belongs to the UTX family.
Contains 1 JmjC domain.
Contains 8 TPR repeats.

Cellular localization Nucleus.

Images



Western blot - Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183)

All lanes : Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183) at 1/1000 dilution ((0.502 µg/ml))

Lane 1 : Wild-type HAP1 whole cell lysate
Lane 2 : KDM6A knockout HAP1 whole cell lysate
Lane 3 : F9 (mouse embryonal carcinoma epithelial cell), whole cell lysate
Lane 4 : THP-1 (human monocytic leukemia monocyte), whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (IRDye® 800CW) (ab216773) and Goat Anti-Mouse IgG H&L (IRDye® 680RD) (ab216776) at 1/20000 dilution

Predicted band size: 154 kDa
Observed band size: 160 kDa

Blocking and diluting buffer and concentration: Intercept® (TBS) Blocking Buffer diluted with an equal volume of 0.1% TBS.

Lanes 1-4: Merged signal (red and green). Green - ab253183 observed at 160 kDa. Red - loading control ab8245 (Mouse monoclonal [6C5] to GAPDH) observed at 36 kDa.

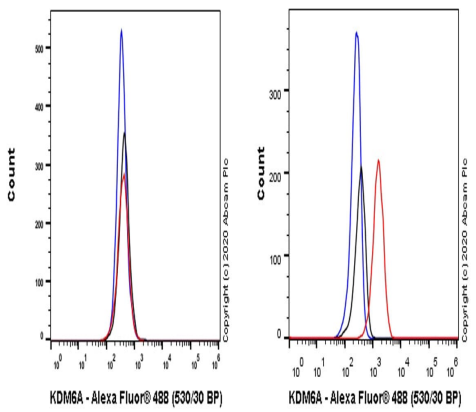
ab253183 Anti-KDM6A antibody [EPR23203-211] was shown to react with KDM6A in HAP1 wild-type cells in Western blot. Loss of signal was observed when KDM6A knockout sample was used. Wild-type and KDM6A knockout samples were subjected to SDS-PAGE.

ab253183 and Anti-GAPDH antibody [6C5] - Loading Control (ab8245) were incubated at room temperature for 2. 5 hours at 1/1000 dilution 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.

Negative control: THP-1 (PMID: 19330029).

The expression profile and molecular weight observed is consistent with what has been described in the literature (PMID: 19330029).

The the bands between 50kDa and 150kDa are caused by degradation based on the result of fresh lysates.

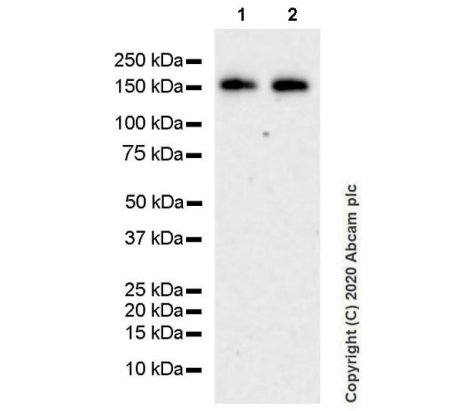


Flow Cytometry (Intracellular) - Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized THP-1 (Human monocytic leukemia monocyte), (Left panel) / HAP1 (Human chronic myelogenous leukemia near-haploid cell line), (Right panel) cells labelling KDM6A / UTX with ab253183 at 1/50 dilution (1µg) (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

Negative control: THP-1.



Western blot - Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183)

All lanes : Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183) at 1/1000 dilution ((0.502 µg/ml))

Lane 1 : HEK-293T (human embryonic kidney epithelial cell), whole cell lysate
Lane 2 : NIH/3T3 (mouse embryonic fibroblast), whole cell lysate

Lysates/proteins at 20 µg per lane.

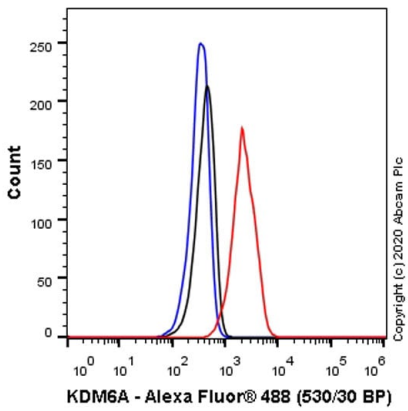
Secondary
All lanes : Goat Anti-Rabbit IgG, (H+L), Peroxidase conjugated (ab97051) at 1/100000 dilution

Predicted band size: 154 kDa
Observed band size: 160 kDa

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

Fresh lysates were used in this WB.

Exposure time: 3 minutes.



Flow Cytometry (Intracellular) - Anti-KDM6A / UTX antibody [EPR23203-211] (ab253183)

Intracellular flow cytometric analysis of 4% paraformaldehyde fixed, 90% methanol permeabilized F9 (Mouse embryonal carcinoma epithelial cell) cells labelling KDM6A / UTX with ab253183 at 1/500 dilution (0.1µg) (Red) compared with a Rabbit monoclonal IgG (ab172730) isotype control (Black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody) (Blue).

A Goat anti rabbit IgG (Alexa Fluor® 488, ab150077) at 1/2000 dilution was used as the secondary antibody.

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