

Product datasheet

Anti-Dnmt1 antibody [EPR18453] ab188453

KO VALIDATED Recombinant RabMAB

★★★★★ 4 Abreviews 2 References 11 Images

Overview

Product name	Anti-Dnmt1 antibody [EPR18453]
Description	Rabbit monoclonal [EPR18453] to Dnmt1
Host species	Rabbit
Tested applications	Suitable for: Flow Cyt, IHC-P, ICC/IF, WB
Species reactivity	Reacts with: Mouse, Rat, Human
Immunogen	Synthetic peptide within Human Dnmt1 aa 1500-1600. The exact sequence is proprietary. Database link: P26358
Positive control	WB: HEK-293, Jurkat, HeLa, Neuro-2a and NIH/3T3 whole cell lysates. IHC-P: Human placenta, mouse stomach and rat stomach tissues. ICC/IF: Wild-type HAP1, HeLa, 293T and Neuro-2a cells.
General notes	Our RabMAB [®] technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to RabMab[®] patents . This product is a recombinant rabbit monoclonal antibody .

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% PBS, 40% Glycerol, 0.05% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR18453
Isotype	IgG

Applications

Our [Abpromise guarantee](#) covers the use of **ab188453** in the following tested applications.

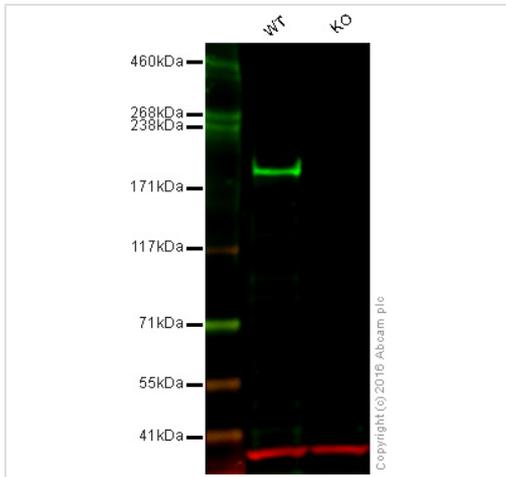
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt		Use at an assay dependent concentration. ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.
IHC-P		1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
ICC/IF	★★★★★	Use a concentration of 1 µg/ml.
WB	★★★★★	1/1000. Detects a band of approximately 183 kDa (predicted molecular weight: 183 kDa).

Target

Function	Methylates CpG residues. Preferentially methylates hemimethylated DNA. Associates with DNA replication sites in S phase maintaining the methylation pattern in the newly synthesized strand, that is essential for epigenetic inheritance. Associates with chromatin during G2 and M phases to maintain DNA methylation independently of replication. It is responsible for maintaining methylation patterns established in development. DNA methylation is coordinated with methylation of histones. Mediates transcriptional repression by direct binding to HDAC2. In association with DNMT3B and via the recruitment of CTCFL/BORIS, involved in activation of BAG1 gene expression by modulating dimethylation of promoter histone H3 at H3K4 and H3K9.
Tissue specificity	Ubiquitous; highly expressed in fetal tissues, heart, kidney, placenta, peripheral blood mononuclear cells, and expressed at lower levels in spleen, lung, brain, small intestine, colon, liver, and skeletal muscle. Isoform 2 is less expressed than isoform 1.
Sequence similarities	Belongs to the C5-methyltransferase family. Contains 2 BAH domains. Contains 1 CXXC-type zinc finger.
Domain	The N-terminal part is required for homodimerization and acts as a regulatory domain.
Post-translational modifications	Sumoylated; sumoylation increases activity.
Cellular localization	Nucleus.

Images



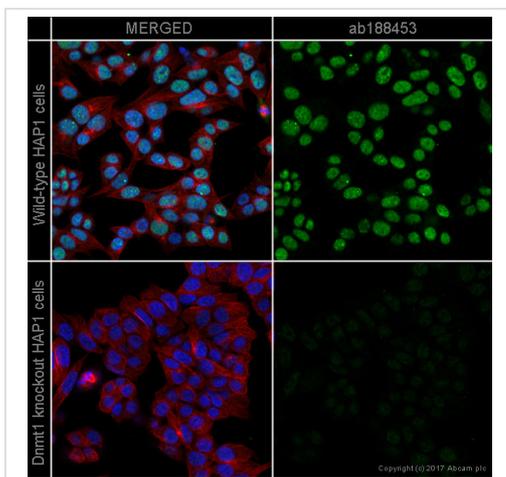
Western blot - Anti-Dnmt1 antibody [EPR18453] (ab188453)

Lane 1: Wild type HAP1 whole cell lysate (20 µg)

Lane 2: DNMT1 knockout HAP1 whole cell lysate (20 µg)

Lanes 1 - 2: Merged signal (red and green). Green - ab188453 observed at 183 kDa. Red - loading control, ab8245, observed at 37 kDa.

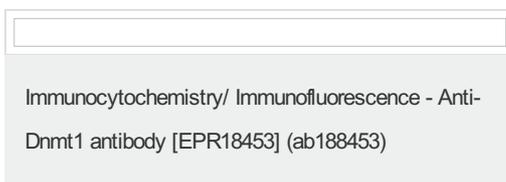
ab188453 was shown to specifically react with DNMT1 when DNMT1 knockout samples were used. Wild-type and DNMT1 knockout samples were subjected to SDS-PAGE. Ab188453 and ab8245 (Mouse anti GAPDH loading control) were incubated overnight at 4°C at a 1/1000 dilution and 1/10000 dilution respectively. Blots 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 1/10000 dilution for 1 hour at room temperature before imaging.



Immunocytochemistry/ Immunofluorescence - Anti-Dnmt1 antibody [EPR18453] (ab188453)

ab188453 staining Dnmt1 in wild-type HAP1 cells (top panel) and DNMT1 knockout HAP1 cells (bottom panel). The cells were fixed with 4% formaldehyde (10min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab188453 at 1 µg/ml and ab195889 at 1/250 dilution (shown in pseudocolour red) overnight at +4°C, followed by a further incubation at room temperature for 1h with a goat secondary antibody to Rabbit IgG (Alexa Fluor® 488) (ab150081) at 2 µg/ml (shown in green). Nuclear DNA was labelled in blue with DAPI.

Image was taken with a confocal microscope (Leica-Microsystems, TCS SP8).



Immunocytochemistry/ Immunofluorescence - Anti-Dnmt1 antibody [EPR18453] (ab188453)

Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Neuro-2a (Mouse neuroblastoma cell line) cells labeling Dnmt1 with ab188453 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488)

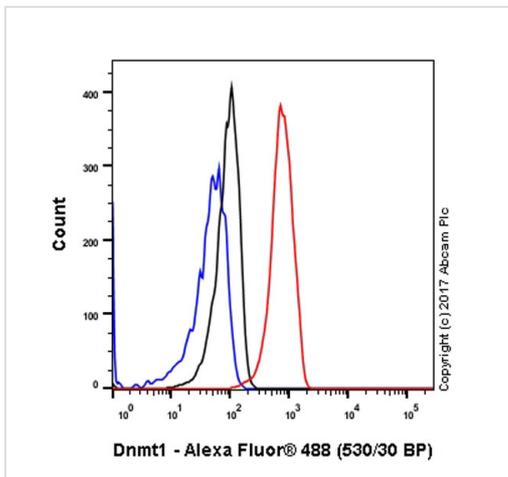
([ab150077](#)) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on Neuro-2a cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb ([ab7291](#)) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) ([ab150120](#)) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

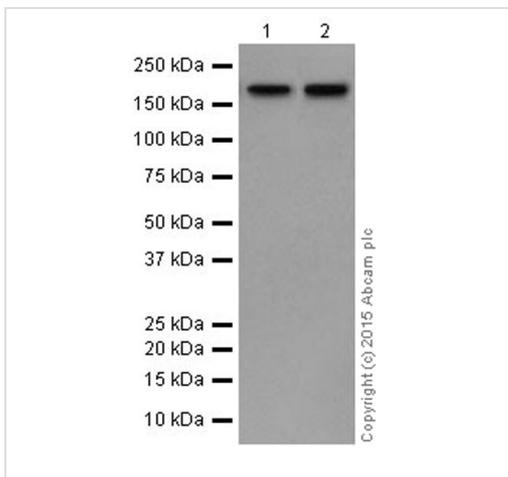
-ve control 1: ab188453 at 1/2000 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) ([ab150120](#)) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb ([ab7291](#)) at 1/1000 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) ([ab150077](#)) secondary antibody at 1/1000 dilution.



Flow Cytometry - Anti-Dnmt1 antibody [EPR18453] (ab188453)

Flow cytometry analysis of HeLa (human cervix adenocarcinoma) cells labeling with purified ab188453 at 1/100 dilution (10ug/ml) (Red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. A Goat anti rabbit IgG (Alexa Fluor® 488)([ab150077](#)) (1/2000 dilution) was used as the secondary antibody. Rabbit monoclonal IgG (Black)([ab172730](#)) was used as a isotype control. Cell without incubation with primary antibody and secondary antibody (Blue) were used as unlabeled control.



Western blot - Anti-Dnmt1 antibody [EPR18453] (ab188453)

All lanes : Anti-Dnmt1 antibody [EPR18453] (ab188453) at 1/1000 dilution

Lane 1 : HEK-293 (Human epithelial cell line from embryonic kidney) whole cell lysate

Lane 2 : Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

Lysates/proteins at 10 µg per lane.

Secondary

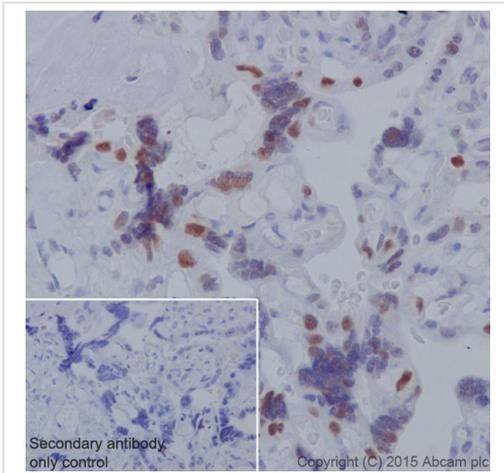
All lanes : Goat Anti-Rabbit IgG H&L (HRP) ([ab97051](#)) at 1/100000 dilution

Predicted band size: 183 kDa

Observed band size: 183 kDa

Exposure time: 15 seconds

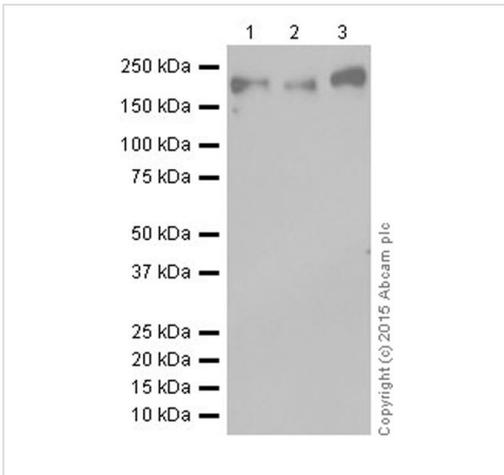
Blocking/Dilution buffer: 5% NFD/MTBST.



Immunohistochemical analysis of paraffin-embedded human placenta tissue labeling Dnmt1 with ab188453 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on human placenta 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.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR18453] (ab188453)



Western blot - Anti-Dnmt1 antibody [EPR18453] (ab188453)

All lanes : Anti-Dnmt1 antibody [EPR18453] (ab188453) at 1/200 dilution

Lane 1 : HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

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

Lane 3 : NIH/3T3 (Mouse embryonic fibroblast cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

Secondary

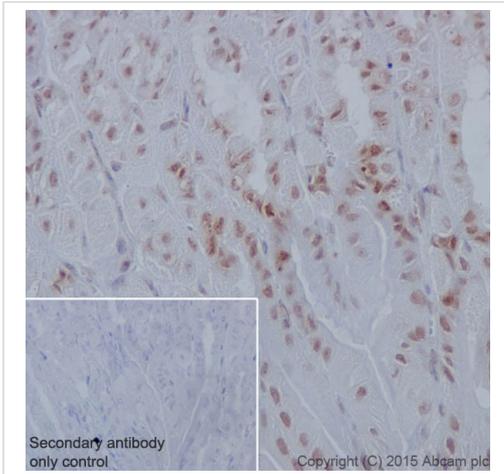
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/100000 dilution

Predicted band size: 183 kDa

Observed band size: 183 kDa

Exposure time: 3 minutes

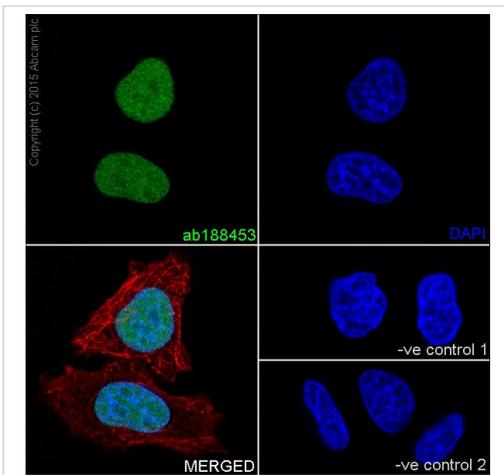
Blocking/Dilution buffer: 5% NFD/MTBST.



Immunohistochemical analysis of paraffin-embedded mouse stomach tissue labeling Dnmt1 with ab188453 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on mouse stomach 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.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR18453] (ab188453)



Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling Dnmt1 with ab188453 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on HeLa cell line. The nuclear counter stain is DAPI (blue).

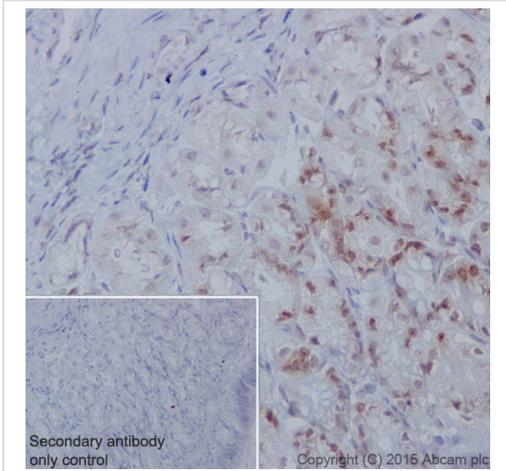
Tubulin is detected with Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (ab150120) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab188453 at 1/2000 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor[®] 594) (ab150120) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor[®] 488) (ab150077) secondary antibody at 1/1000 dilution.

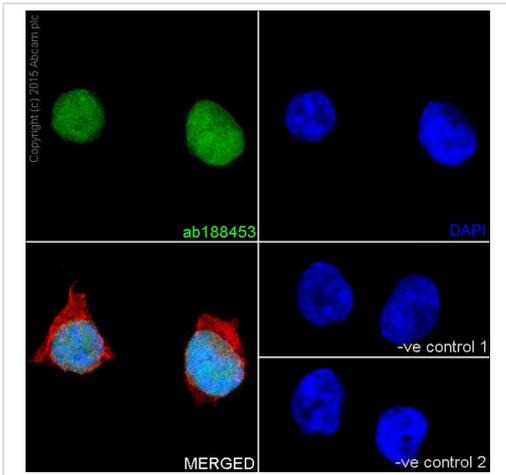
Immunocytochemistry/ Immunofluorescence - Anti-Dnmt1 antibody [EPR18453] (ab188453)



Immunohistochemical analysis of paraffin-embedded rat stomach tissue labeling Dnmt1 with ab188453 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on rat stomach 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.

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR18453] (ab188453)



Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HEK-293 (Human epithelial cell line from embryonic kidney) cells labeling Dnmt1 with ab188453 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on HEK-293 cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (ab7291) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab188453 at 1/2000 dilution followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution.

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Immunocytochemistry/ Immunofluorescence - Anti-Dnmt1 antibody [EPR18453] (ab188453)

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