abcam

# ab134148

# Anti-Dnmt1 antibody [EPR3521(2)]

Recombinant | RabMAb | KO Validated | 20ul selling size

## Key facts

| | |
|---|---|
| **Isotype** | IgG |
| **Host species** | Rabbit |
| **Storage buffer** | pH: 7.2 - 7.4 Preservative: 0.01% Sodium azide Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA |
| **Form** | Liquid |
| **Clonality** | Monoclonal |
| **Immunogen** | The exact immunogen used to generate this antibody is proprietary information. |
| **Clone number** | EPR3521(2) |
| **Purification technique** | Affinity purification Protein A |
| **Concentration** | 0.084 mg/mL The concentration of this product may be batch-dependent<br>Batch concentration finder → |

## Reactivity data

## IHC-P

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/250 - 1/700 |
| **Notes** | Perform heat-mediated antigen retrieval before commencing with IHC staining protocol. |

## WB

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/1000 - 1/10000 |
| **Notes** | - |

## ICC/IF

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/250 |
| **Notes** | - |

## Flow Cyt (Intra)

### Tested

| | |
|---|---|
| **Species** | Human |
| **Dilution info** | 1/20 |
| **Notes** | ab172730 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. |

# Target data

See full target information DNMT1

**Function** DNA methyltransferase that methylates CpG residues (PubMed:17200670, PubMed:18754681, PubMed:21745816, PubMed:26070743). Preferentially methylates hemimethylated DNA (PubMed:21745816, PubMed:26070743). Associates with DNA replication sites in S phase maintaining the methylation pattern in the newly synthesized strand, that is essential for epigenetic inheritance (PubMed:17200670, PubMed:21745816). Associates with chromatin during G2 and M phases to maintain DNA methylation independently of replication (PubMed:21745816). It is responsible for maintaining methylation patterns established in development (PubMed:21745816). DNA methylation is coordinated with methylation of histones (PubMed:16357870). Mediates transcriptional repression by direct binding to HDAC2 (PubMed:10888872). 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 (PubMed:18413740). Probably forms a corepressor complex required for activated KRAS-mediated promoter hypermethylation and transcriptional silencing of tumor suppressor genes (TSGs) or other tumor-related genes in colorectal cancer (CRC) cells (PubMed:24623306). Also required to maintain a transcriptionally repressive state of genes in undifferentiated embryonic stem cells (ESCs) (PubMed:24623306). Associates at promoter regions of tumor suppressor genes (TSGs) leading to their gene silencing (PubMed:24623306).

## Storage

| | |
|---|---|
| **Shipped at conditions** | Conditional Ambient |
| **Appropriate short-term storage duration** | 1-2 weeks |
| **Appropriate short-term storage conditions** | +4°C |
| **Appropriate long-term storage conditions** | -20°C |
| **Aliquoting information** | Upon delivery aliquot |
| **Storage information** | Avoid freeze / thaw cycle |

## Notes

**Species reactivity**
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

**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.

## 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.

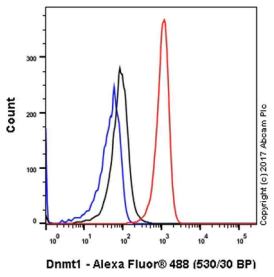
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We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

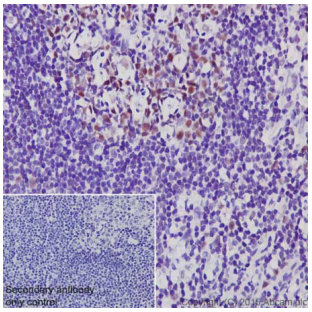
## 9 product images




### Flow Cytometry (Intracellular) - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Flow Cytometry (Intracellular) staining using rabbit Anti-Dnmt1 antibody

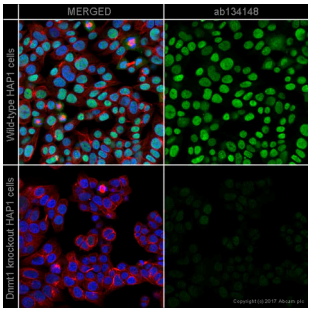
Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling Dnmt1 (red) with unpurified ab134148 at a 1/20 dilution (10ug/mL). Cells were fixed with 4% paraformaldehyde and permeabilized with 90% methanol. A goat anti rabbit IgG (Alexa Fluor® 488) (ab150077) was used as the secondary antibody at a 1/2000 dilution. Black - Rabbit monoclonal IgG (Black) (ab172730). Blue (unlabeled control) - Cell without incubation with primary antibody and secondary antibody (Blue).



### Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Dnmt1 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling Dnmt1 with purified ab134148 at 1/700. Heat mediated antigen retrieval was performed using Tris/EDTA buffer pH 9. ab97051, a HRP-conjugated goat anti-rabbit IgG (H+L) was used as the secondary antibody (1/500). Negative control using PBS instead of primary antibody. Counterstained with hematoxylin.

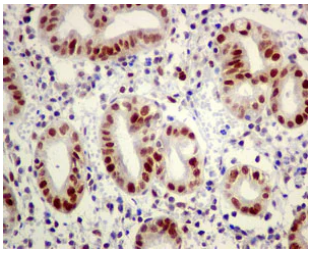



## Immunocytochemistry/ Immunofluorescence - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-Dnmt1 antibody

ab134148 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 ab134148 at 1/250 dilution 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).

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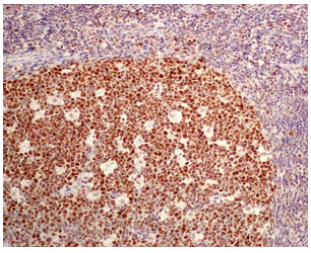


## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Dnmt1 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human stomach tissue labelling Dnmt1 with unpurified ab134148 at a dilution of 1/250.
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

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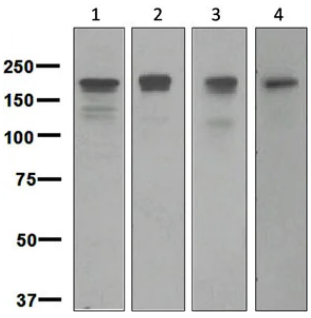


## Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-Dnmt1 antibody

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of human tonsil tissue labelling Dnmt1 with unpurified ab134148 at a dilution of 1/250.
Perform heat mediated antigen retrieval before commencing with IHC staining protocol.

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## Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

All lanes:
Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148) at 1/1000 dilution

Lane 1:
Jurkat cell lysates at 10 µg

Lane 2:
293T cell lysates at 10 µg

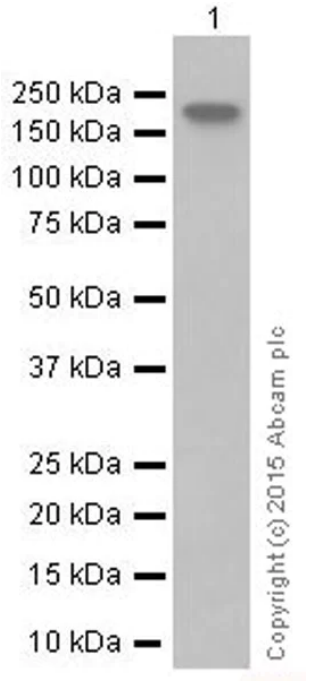
Lane 3:
HeLa cell lysates at 10 µg

Lane 4:
HuT-78 cell lysates at 10 µg

Secondary

All lanes:
HRP-conjugated goat anti-rabbit IgG at 1/2000 dilution

Predicted band size: 183 kDa




## Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Western blot staining of Jurkat whole cell lysate using rabbit Anti-Dnmt1 antibody

Blocking and dilution buffer: 5% NFDM /TBST.
All lanes:
Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148) at 1/5000 dilution

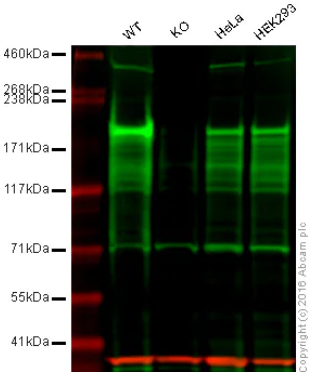
All lanes:
Jurkat whole cell lysate at 10 µg

Secondary

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

Predicted band size: 183 kDa

Observed band size: 183 kDa




## Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

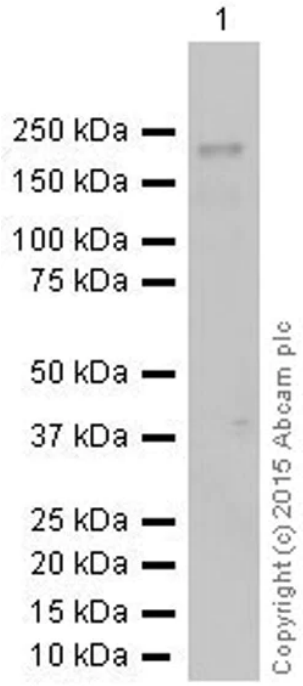
Dnmt1 Western blot staining using rabbit Anti-Dnmt1 antibody

**Lane 1:** Wild type HAP1 whole cell lysate (20 µg)
**Lane 2:** DNMT1 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 - ab134148 observed at 183 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab134148 was shown to recognize DNMT1 when DNMT1 knockout samples were used, along with additional cross-reactive bands. Wild-type and DNMT1 knockout samples were subjected to SDS-PAGE. ab134148 and ab8245 (Mouse anti-GAPDH loading control) were incubated overnight at 4°C at 1/1000 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.

All lanes:
Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Predicted band size: 183 kDa




# Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148)

Dnmt1 Western blot staining of 293T whole cell lysate using rabbit Anti-Dnmt1 antibody

Blocking and dilution buffer: 5% NFDM /TBST.
All lanes:
Western blot - Anti-Dnmt1 antibody [EPR3521(2)] (ab134148) at 1/5000 dilution

All lanes:
293T whole cell lysate at 10 µg

Secondary

All lanes:
Western blot - Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 183 kDa

Observed band size: 183 kDa

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