Anti-DNMT1 Antibody [A3A8-R]

HA601281

Product Type:	Recombinant Mouse monoclonal IgG1, primary antibodies
Species reactivity:	Human
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 183 kDa
Clone number:	A3A8-R

Description: 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. 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. Also required to maintain a transcriptionally repressive state of genes in undifferentiated embryonic stem cells (ESCs). Associates at promoter regions of tumor suppressor genes (TSGs) leading to their gene silencing. Promotes tumor growth.

Immunogen: Synthetic peptide within Human Dnmt1 aa 500-600 / 1616.

Positive control: HeLa cell lysate, 293T cell lysate, HepG2 cell lysate, Jurkat cell lysate, HeLa, human testis tissue.

Subcellular location: Nucleus.

Database links: SwissProt: P26358 Human

Recommended Dilutions:

WB	1:1,000
IF-Cell	1:100
IHC-P	1:800

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). Store at -20℃ long term.

Purity: Protein A affinity purified.

Images

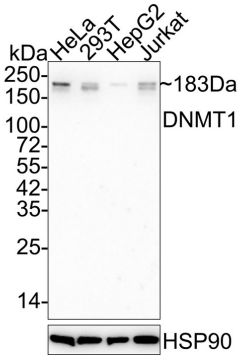

Fig1: Western blot analysis of DNMT1 on different lysates with Mouse anti-DNMT1 antibody (HA601281) at 1/1,000 dilution.

Lane 1: HeLa cell lysate, 20 µg/Lane
Lane 2: 293T cell lysate, 20 µg/Lane
Lane 3: HepG2 cell lysate, 20 µg/Lane
Lane 4: Jurkat cell lysate, 20 µg/Lane

Predicted band size: 183 kDa
Observed band size: 183 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA601281) at 1/1,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Mouse IgG - HRP Secondary Antibody (HA1006) at 1/50,000 dilution was used for 1 hour at room temperature.

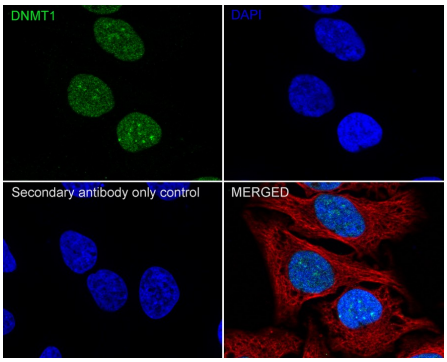

Fig2: Immunocytochemistry analysis of HeLa cells labeling DNMT1 with Mouse anti-DNMT1 antibody (HA601281) at 1/100 dilution.

Cells were fixed in 4% paraformaldehyde for 20 minutes at room temperature, permeabilized with 0.1% Triton X-100 in PBS for 5 minutes at room temperature, then blocked with 1% BSA in 10% negative goat serum for 1 hour at room temperature. Cells were then incubated with Mouse anti-DNMT1 antibody (HA601281) at 1/100 dilution in 1% BSA in PBST overnight at 4 ℃. Goat Anti-Mouse IgG H&L (iFluor™ 488, HA1125) was used as the secondary antibody at 1/1,000 dilution. PBS instead of the primary antibody was used as the secondary antibody only control. Nuclear DNA was labelled in blue with DAPI.

beta Tubulin (ET1602-4, red) was stained at 1/100 dilution overnight at +4℃. Goat Anti-Rabbit IgG H&L (iFluor™ 594, HA1122) were used as the secondary antibody at 1/1,000 dilution.

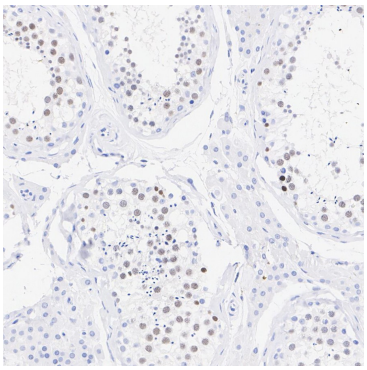

Fig3: Immunohistochemical analysis of paraffin-embedded human testis tissue with Mouse anti-DNMT1 antibody (HA601281) at 1/800 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) (high pressure) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH_2O and PBS, and then probed with the primary antibody (HA601281) at 1/800 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

Background References

1. Li T. et. al. Structural and mechanistic insights into UHRF1-mediated DNMT1 activation in the maintenance DNA methylation. Nucleic Acids Res. 2018 Apr 6;46(6):3218-3231.
2. Du WW. et. al. A circular RNA circ-DNMT1 enhances breast cancer progression by activating autophagy. Oncogene. 2018 Nov;37(44):5829-5842.