Anti-MLH1 Antibody [PD01-61]

HA721325

Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IHC-P
Molecular Wt:	Predicted band size: 85 kDa
Clone number:	PD01-61

Description: DNA-mismatch repair (MMR) is an essential process in maintaining genetic stability. Lack of a functional DNA-mismatch repair pathway is a common characteristic of several different types of human cancers, either due to an MMR gene mutation or promoter methylation gene silencing. MLH1 is an integral part of the protein complex responsible for mismatch repair and is expressed in lymphocytes, heart, colon, breast, lung, spleen, testis, prostate, thyroid and gall bladder tissues, and is methylated in several ovarian tumors. Loss of MLH1 protein expression is associated with a mutated phenotype, microsatellite instability and a predisposition to cancer. In hereditary nonpolyposis colorectal cancer (HNPCC), an autosomal dominant inherited cancer syndrome that signifies a high risk of colorectal and various other types of cancer, the MLH1 gene exhibits a pathogenic mutation. Certain cancer cell lines, including leukemia CCRF-CEM, colon HCT 116 and KM12, and ovarian cancers SK-OV-3 and IGROV-1, show complete deficiency of MLH1, while MLH1 is expressed in 60% of melanomas, 70% of noninvasive squamous cell carcinomas and 30% of invasive squamous cell carcinomas.

Immunogen: Synthetic peptide corresponding to Human MLH1 aa 700-756.

Positive control: HeLa cell lysate, HCT 116 cell lysate, A549 cell lysate, C2C12 cell lysate, NIH/3T3 cell lysate, human appendix tissue, human colon tissue, human B-cell lymphoblastic lymphoma tissue, human NKT cell lymphoma tissue, mouse testis tissue, mouse thymus tissue, human colon carcinoma tissue, rat small intestine tissue.

Subcellular location: Nucleus.

Database links: SwissProt: P40692 Human | Q9JK91 Mouse | P97679 Rat

Recommended Dilutions:

WB	1:2,000
IHC-P	1:200-1:1,000

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

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Images

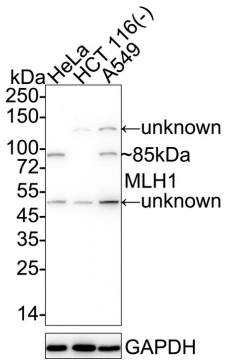

Fig1: Western blot analysis of MLH1 on different lysates with Rabbit anti-MLH1 antibody (HA721325) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HCT 116 cell lysate (negative)
Lane 3: A549 cell lysate

Lysates/proteins at 15 µg/Lane.

Predicted band size: 85 kDa
Observed band size: 85 kDa

Exposure time: 2 minutes; ECL: K1801;

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 (HA721325) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

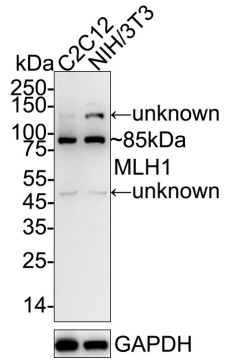

Fig2: Western blot analysis of MLH1 on different lysates with Rabbit anti-MLH1 antibody (HA721325) at 1/2,000 dilution.

Lane 1: C2C12 cell lysate
Lane 2: NIH/3T3 cell lysate

Lysates/proteins at 30 µg/Lane.

Predicted band size: 85 kDa
Observed band size: 85 kDa

Exposure time: 1 minute; ECL: K1801;

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 (HA721325) at 1/2,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

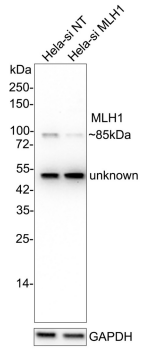

Fig3: Western blot analysis of MLH1 on different lysates with Rabbit anti-MLH1 antibody (HA721325) at 1/2,000 dilution.

Lane 1: Hela-si NT cell lysate
Lane 2: Hela-si MLH1 cell lysate

Lysates/proteins at 10 µg/Lane.

Predicted band size: 85 kDa
Observed band size: 85 kDa

Exposure time: 3 minutes;

4-20% SDS-PAGE gel.

HA721325 was shown to specifically react with MLH1 in Hela-si NT cells. Weakened band was observed when Hela-si MLH1 sample was tested. Hela-si NT and Hela-si MLH1 samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (HA721325, 1/2,000) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

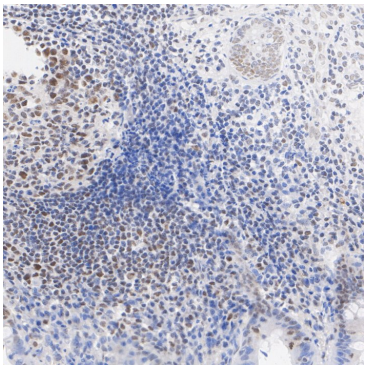

Fig4: Immunohistochemical analysis of paraffin-embedded human appendix tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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.

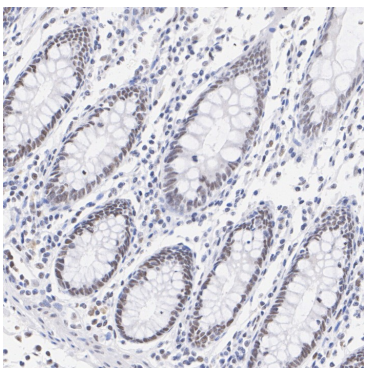

Fig5: Immunohistochemical analysis of paraffin-embedded human colon tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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.

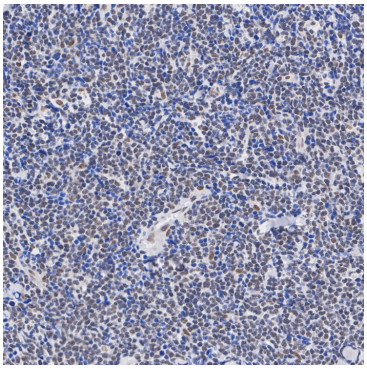

Fig6: Immunohistochemical analysis of paraffin-embedded human B-cell lymphoblastic lymphoma tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/200 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.

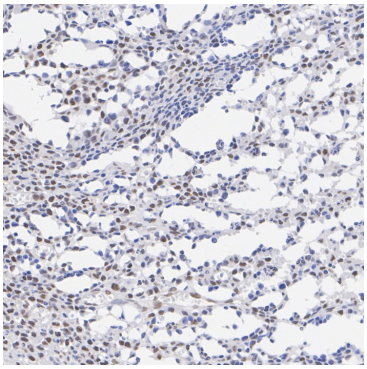

Fig7: Immunohistochemical analysis of paraffin-embedded human NKT cell lymphoma tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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.

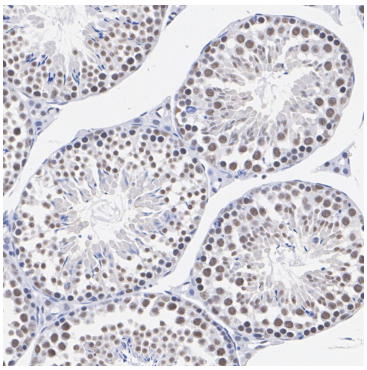

Fig8: Immunohistochemical analysis of paraffin-embedded mouse testis tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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.

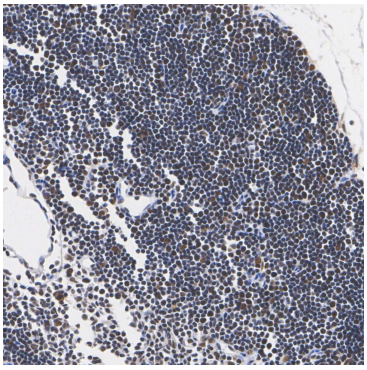

Fig9: Immunohistochemical analysis of paraffin-embedded mouse thymus tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/200 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/200 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.

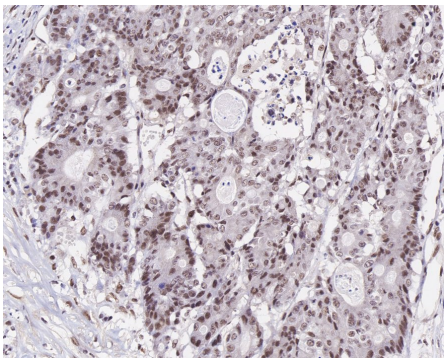

Fig10: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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.

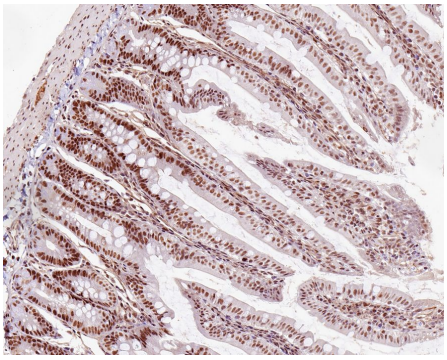

Fig11: Immunohistochemical analysis of paraffin-embedded rat small intestine tissue with Rabbit anti-MLH1 antibody (HA721325) at 1/1,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) 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 (HA721325) at 1/1,000 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. Guan J et al. MLH1 Deficiency-Triggered DNA Hyperexcision by Exonuclease 1 Activates the cGAS-STING Pathway. Cancer Cell. 2021 Jan
2. Cannavo E et al. Regulation of the MLH1-MLH3 endonuclease in meiosis. Nature. 2020 Oct