

Anti-MLH1 Antibody [PSH01-74]

HA721726



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human
Applications:	IHC, IF-Cell, IF-Tissue
Molecular Wt:	Predicted band size: 85 kDa
Clone number:	PSH01-74

Description: Heterodimerizes with PMS2 to form MutL alpha, a component of the post-replicative DNA mismatch repair system (MMR). DNA repair is initiated by MutS alpha (MSH2-MSH6) or MutS beta (MSH2-MSH3) binding to a dsDNA mismatch, then MutL alpha is recruited to the heteroduplex. Assembly of the MutL-MutS-heteroduplex ternary complex in presence of RFC and PCNA is sufficient to activate endonuclease activity of PMS2. It introduces single-strand breaks near the mismatch and thus generates new entry points for the exonuclease EXO1 to degrade the strand containing the mismatch. DNA methylation would prevent cleavage and therefore assure that only the newly mutated DNA strand is going to be corrected. MutL alpha (MLH1-PMS2) interacts physically with the clamp loader subunits of DNA polymerase III, suggesting that it may play a role to recruit the DNA polymerase III to the site of the MMR. Also implicated in DNA damage signaling, a process which induces cell cycle arrest and can lead to apoptosis in case of major DNA damages. Heterodimerizes with MLH3 to form MutL gamma which plays a role in meiosis.

Immunogen:	Recombinant protein within human MLH1 aa 351-550 / 756.
Positive control:	Human appendix tissue, human colon cancer tissue, human tonsil tissue, A431, HeLa.
Subcellular location:	Nucleus, Chromosome.
Database links:	SwissProt: P40692 Human
Recommended Dilutions:	
IHC	1:400
IF-Cell	1:100
IF-Tissue	1:50
Storage Buffer:	PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.
Storage Instruction:	Store at +4°C after thawing. Aliquot store at -20°C. Avoid repeated freeze / thaw cycles.
Purity:	Protein A affinity purified.

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Technical:0086-571-89986345

Service mail:support@huabio.cn

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Images

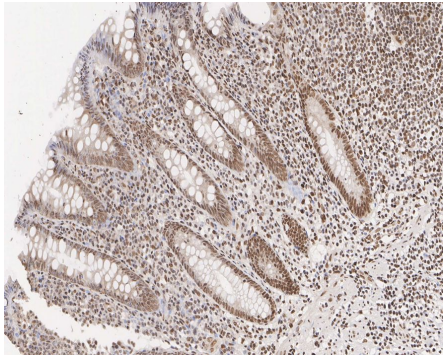


Fig1: Immunohistochemical analysis of paraffin-embedded human appendix tissue with Rabbit anti-MLH1 antibody (HA721726) at 1/400 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA721726) at 1/400 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.

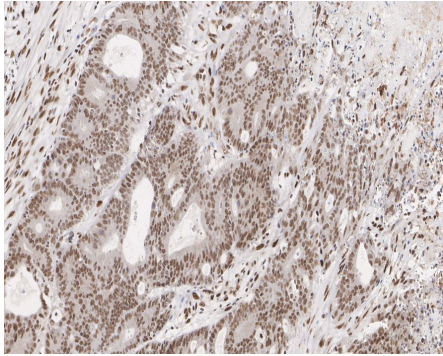


Fig2: Immunohistochemical analysis of paraffin-embedded human colon cancer tissue with Rabbit anti-MLH1 antibody (HA721726) at 1/400 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA721726) at 1/400 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.

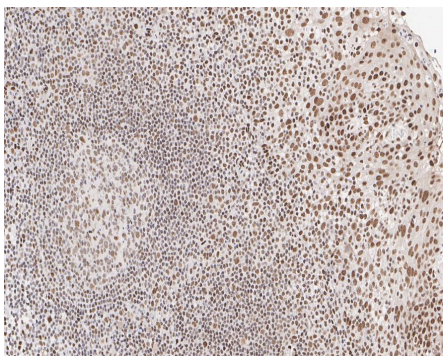
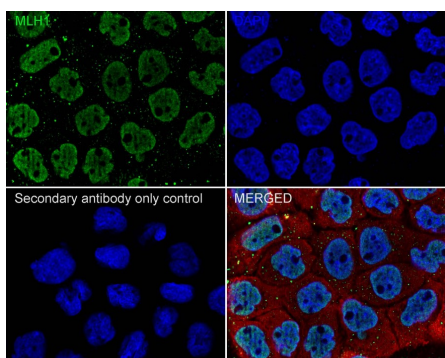


Fig3: Immunohistochemical analysis of paraffin-embedded human tonsil tissue with Rabbit anti-MLH1 antibody (HA721726) at 1/400 dilution.

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 ddH₂O and PBS, and then probed with the primary antibody (HA721726) at 1/400 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.

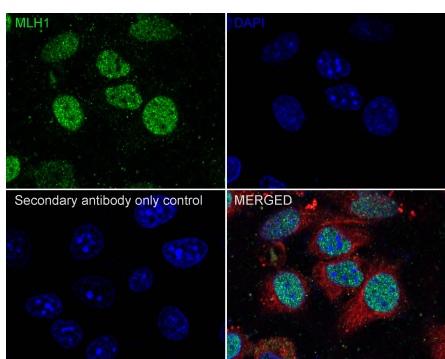
Fig4: Immunocytochemistry analysis of A431 cells labeling MLH1 with Rabbit anti-MLH1 antibody (HA721726) at 1/100 dilution.



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 Rabbit anti-MLH1 antibody (HA721726) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) 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 (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

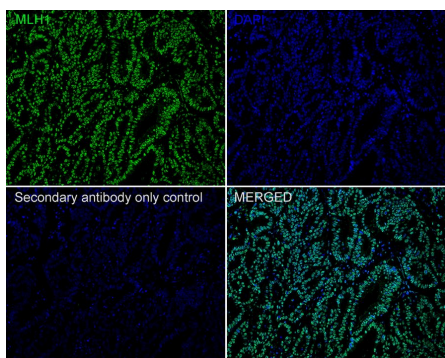
Fig5: Immunocytochemistry analysis of HeLa cells labeling MLH1 with Rabbit anti-MLH1 antibody (HA721726) at 1/100 dilution.



Cells were fixed in 100% precooled methanol 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 Rabbit anti-MLH1 antibody (HA721726) at 1/100 dilution in 1% BSA in PBST overnight at 4 °C. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) 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 (M1305-2, red) was stained at 1/100 dilution overnight at +4°C. Goat Anti-Mouse IgG H&L (iFluor™ 594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Fig6: Immunofluorescence analysis of paraffin-embedded human colon cancer tissue labeling MLH1 with Rabbit anti-MLH1 antibody (HA721726) at 1/50 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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA721726, green) at 1/50 dilution overnight at 4 °C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

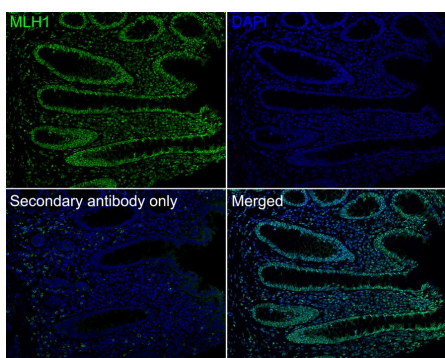


Fig7: Immunofluorescence analysis of paraffin-embedded human appendix tissue labeling MLH1 with Rabbit anti-MLH1 antibody (HA721726) at 1/50 dilution.

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 10% negative goat serum for 1 hour at room temperature, washed with PBS, and then probed with the primary antibody (HA721726, green) at 1/50 dilution overnight at 4 °C, washed with PBS. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclei were counterstained with DAPI (blue).

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

Background References

1. Manzoor S, Saber-Ayad M, Maghazachi AA, Hamid Q, Muhammad JS. MLH1 mediates cytoprotective nucleophagy to resist 5-Fluorouracil-induced cell death in colorectal carcinoma. *Neoplasia*. 2022 Feb;24(2):76-85. doi: 10.1016/j.neo.2021.12.003. Epub 2021 Dec 21. PMID: 34952246; PMCID: PMC8695220.

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