abcam

ab251498

Anti-MSH6 antibody [EPR20316] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR20316
Purification technique	Affinity purification Protein A
Specificity	This antibody recognizes isoforms 1 and 4 of MSH6.
Concentration	1.018 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

IP

Tested

Species	Human

Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

ICC/IF

Tested

Species	Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	-

IHC-P

Tested

Species	Human
Dilution info	-
Notes	-

Target data

See full target information MSH6

Function	Component of the post-replicative DNA mismatch repair system (MMR). Heterodimerizes with MSH2 to form MutS alpha, which binds to DNA mismatches thereby initiating DNA repair. When bound, MutS alpha bends the DNA helix and shields approximately 20 base pairs, and recognizes single base mismatches and dinucleotide insertion-deletion loops (IDL) in the DNA. After mismatch binding, forms a ternary complex with the MutL alpha heterodimer, which is thought to be responsible for directing the downstream MMR events, including strand discrimination, excision, and resynthesis. ATP binding and hydrolysis play a pivotal role in mismatch repair functions. The ATPase activity associated with MutS alpha regulates binding similar to a molecular switch: mismatched DNA provokes ADP-->ATP exchange, resulting in a discernible conformational transition that converts MutS alpha into a sliding clamp capable of hydrolysis-independent diffusion along the DNA backbone. This transition is crucial for mismatch repair. MutS alpha may also play a role in DNA homologous recombination repair. Recruited on chromatin in G1 and early S phase via its PWWP domain that specifically binds trimethylated 'Lys-36' of histone H3 (H3K36me3): early recruitment to chromatin to be replicated allowing a quick identification of mismatch repair to initiate the DNA mismatch repair reaction.

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab251498 is the carrier-free version of ab208940.

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.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	MSH6 also known as MutS Homolog 6 is a DNA repair protein that plays a role in the mismatch repair (MMR) system. It has a molecular mass of approximately 136 kDa. MSH6 forms a heterodimer with MSH2 called MutSα and this complex identifies base-pair mismatches and insertion-deletion loops during DNA replication. It is expressed in various tissues throughout the body and high levels are often found in proliferative tissues where active DNA replication occurs.
Biological function summary	MSH6 functions as part of the MMR complex which is essential for maintaining genomic stability. The MutSα complex where MSH6 pairs with MSH2 operates along with other proteins in the MMR pathway to correct DNA replication errors. MSH6 is also known to interact with PCNA a DNA polymerase processivity factor which facilitates its role in the repair process.
Pathways	MSH6 participates prominently in the DNA mismatch repair pathway. This pathway is critical for correcting DNA errors and preventing mutations during replication. In association with MLH1-PMS2 (MutLα complex) MSH6 ensures that DNA integrity is preserved. Additionally MSH6 is involved in the base excision repair (BER) pathway where it collaborates with other repair proteins to fix small base lesions.
Associated diseases and disorders	MSH6 has a significant connection with Lynch syndrome also known as hereditary nonpolyposis colorectal cancer (HNPCC). This condition is characterized by germline mutations in MMR genes including MSH6 leading to increased cancer risk particularly in the colon. Moreover alterations in MSH6 can contribute to microsatellite instability a feature seen in certain types of endometrial cancer. Mutations in MSH2 often accompany MSH6 mutations in these disorders further impacting the MMR pathway's efficiency.

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.

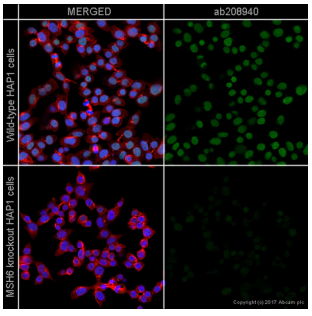
We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

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.

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9 product images

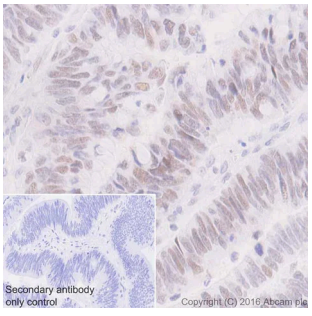


Immunocytochemistry/ Immunofluorescence - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

MSH6 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
ab208940 staining MSH6 in wild-type HAP1 cells (top panel) and MSH6 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 ab208940 at 1µg/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).



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

MSH6 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.

Immunohistochemical analysis of paraffin-embedded Human colon cancer tissue labeling MSH6 with ab208940 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Nuclear staining on tumor cells of human colon cancer is observed [PMID: 26315971].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

MSH6 Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) staining using rabbit Anti-MSH6 antibody

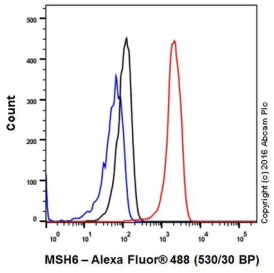
This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Immunohistochemical analysis of paraffin-embedded Human endometrium tissue labeling MSH6 with ab208940 at 1/2000 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

Nuclear staining on tumor cells of human endometrium is observed [PMID: 8942985].

Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit IgG H&L (HRP) Ready to use.

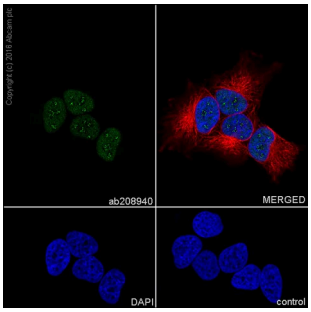
Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.



Flow Cytometry (Intracellular) - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

MSH6 Flow Cytometry (Intracellular) staining using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling MSH6 with ab208940 at 1/500 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730; black) and an unlabeled control (cells without incubation with primary antibody and secondary antibody; blue). Goat anti rabbit IgG (Alexa Fluor® 488) at 1/2000 dilution was used as the secondary antibody.



Immunocytochemistry/ Immunofluorescence - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

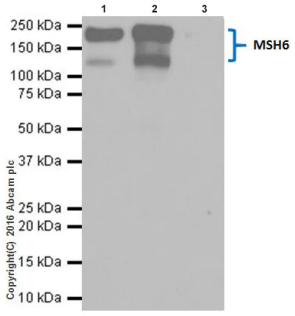
MSH6 Immunocytochemistry/ Immunofluorescence staining using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Immunofluorescent analysis of 4% paraformaldehyde-fixed HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling MSH6 with ab208940 at 1/1000 dilution, followed by Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) secondary antibody at 1/1000 dilution (green).

Confocal image showing nuclear staining on HeLa cell line.

The nuclear counterstain is DAPI (blue). Tubulin is detected with ab195889 (Anti-alpha Tubulin antibody [DM1A] - Microtubule Marker (Alexa Fluor® 594)) at 1/200 dilution (red).

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat anti-rabbit IgG (Alexa Fluor® 488) (ab150077) at 1/1000 dilution.



Immunoprecipitation - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
MSH6 was immunoprecipitated from 0.35 mg of HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate with ab208940 at 1/40 dilution.

Western blot was performed from the immunoprecipitate using ab208940 at 1/1000 dilution.

VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/1000 dilution.

Lane 1: HeLa whole cell lysate, 10 µg (Input).

Lane 2: ab208940 IP in HeLa whole cell lysate.

Lane 3: Rabbit monoclonal IgG (ab172730) instead of ab208940 in HeLa whole cell lysate.

Blocking and dilution buffer: 5% NFDM/TBST.

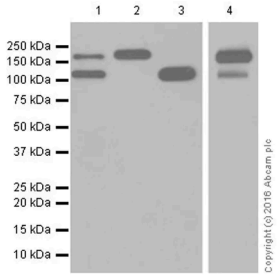
Exposure time: 8 seconds.

The observed molecular weight corresponds to isoform 1 (160kDa) and isoform 4 (120kDa) which is consistent with the Uniprot annotation.

All lanes:
Immunoprecipitation - Anti-MSH6 antibody [EPR20316] (ab208940)

Predicted band size: 153 kDa

Observed band size: 120 kDa, 160 kDa



Western blot - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

Exposure time: Lanes 1-3: 3 minutes; Lane 4: 15 seconds.

This antibody recognizes isoforms 1 and 4 of MSH6. The observed molecular weight corresponds to isoform 1 (160kDa) and isoform 4 (120kDa) which is consistent with the Uniprot annotation.

All lanes:
Western blot - Anti-MSH6 antibody [EPR20316] (ab208940) at 1/1000 dilution

Lane 1:
A431 (Human epidermoid carcinoma cell line) whole cell lysate at 20 µg

Lane 2:
SW480 (Human colorectal adenocarcinoma cell line) whole cell lysate at 20 µg

Lane 3:
HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate at 20 µg

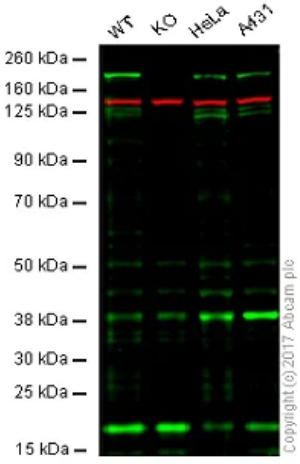
Lane 4:
HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate at 20 µg

Secondary

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

Predicted band size: 153 kDa

Observed band size: 120 kDa, 160 kDa



Western blot - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)

MSH6 Western blot staining using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Lane 1: Wild type HAP1 whole cell lysate (20 µg)

Lane 2: MSH6 knockout HAP1 whole cell lysate (20 µg)

Lane 3: HeLa whole cell lysate (20 µg)

Lane 4: A431 whole cell lysate (20 µg)

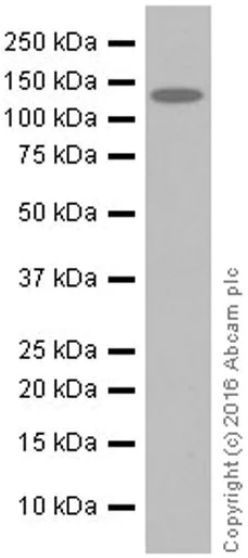
Lanes 1 - 4: Merged signal (red and green). Green - ab208940 observed at 165 kDa. Red - loading control, ab18058, observed at 130 kDa.

ab208940 was shown to specifically react with MSH6 in wild-type HAP1 cells along with additional cross-reactive bands. No band was observed when MSH6 knockout samples were used. Wild-type and MSH6 knockout samples were subjected to SDS-PAGE. ab208940 and ab18058 (Mouse anti Vinculin loading control) were incubated overnight at 4°C at 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 1/20000 dilution for 1 hour at room temperature before imaging.
All lanes:
Western blot - Anti-MSH6 antibody [EPR20316] (ab208940)

Predicted band size: 153 kDa

Western blot - Anti-MSH6 antibody [EPR20316] - BSA and Azide free (ab251498)



MSH6 Western blot staining of Human testis tissue lysate using rabbit Anti-MSH6 antibody

This data was developed using ab208940, the same antibody clone in a different buffer formulation.
Blocking and dilution buffer: 5% NFDM/TBST.

All lanes:
Western blot - Anti-MSH6 antibody [EPR20316] (ab208940) at 1/1000 dilution

All lanes:
Human testis tissue lysate at 20 µg

Secondary

All lanes:
Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/10000 dilution

Predicted band size: 153 kDa

Observed band size: 120 kDa

Exposure time: 3min

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