abcam

Product datasheet

Anti-MSH6 antibody [SP93] ab99889



重组 RabMAb

1 References 10 图像

概述

产品名称 Anti-MSH6抗体[SP93]

描述 兔单克隆抗体[SP93] to MSH6

宿主 Rabbit

经测试应用 适用于: WB, ICC/IF, IHC-P, Flow Cyt (Intra)

种属反应性 与反应: Mouse, Human

免疫原 Synthetic peptide within Human MSH6 aa 350-450 (internal sequence). The exact sequence is

proprietary.

Database link: P52701

表位 Internal region

阳性对照 IHC-P: Human rectal carcinoma, colon, and colon carcinoma tissues; Mouse colon Tissue ICC/IF:

HeLa cells, HAP1 cells (HAP1-MSH6 knockout cell line used as negative cell line). Flow Cyt

(Intra): HeLa cells.

常规说明 To see more of the key markers and tools you need to study the hallmarks of cancer, including

genome instability and mutation, please visit the following page.

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 production

For more information see here.

This product is FOR RESEARCH USE ONLY. For commercial use, please contact

partnerships@abcam.com.

性能

形式 Liquid

存放说明 Shipped at 4°C. Store at +4°C.

存储溶液 pH: 7.60

> Preservative: 0.1% Sodium azide Constituents: PBS, 1% BSA

纯**度** Protein A/G purified

纯**化说明** Purified from TCS by protein A/G.

 克隆
 单克隆

 克隆编号
 SP93

 同种型
 IqG

应用

The Abpromise guarantee Abpromise™承诺保证使用ab99889于以下的经测试应用

"应用说明"部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration.
ICC/IF		1/250.
IHC-P		1/100. Antigen retrieval: boil tissue section in 1 mM EDTA, pH 8.0 for 10 minutes followed by cooling at room temperature for 20 minutes. Primary antibody incubation: 30 minutes at room temperature.
Flow Cyt (Intra)		1/20.

靶标

功能

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.

疾病相关

Defects in MSH6 are the cause of hereditary non-polyposis colorectal cancer type 5 (HNPCC5) [MIM:600678]. Mutations in more than one gene locus can be involved alone or in combination in the production of the HNPCC phenotype (also called Lynch syndrome). Most families with clinically recognized HNPCC have mutations in either MLH1 or MSH2 genes. HNPCC is an autosomal, dominantly inherited disease associated with marked increase in cancer susceptibility. It is characterized by a familial predisposition to early onset colorectal carcinoma (CRC) and extra-colonic cancers of the gastrointestinal, urological and female reproductive tracts. HNPCC is reported to be the most common form of inherited colorectal cancer in the Western world. Cancers in HNPCC originate within benign neoplastic polyps termed adenomas. Clinically, HNPCC is often divided into two subgroups. Type I: hereditary predisposition to colorectal

cancer, a young age of onset, and carcinoma observed in the proximal colon. Type II: patients have an increased risk for cancers in certain tissues such as the uterus, ovary, breast, stomach, small intestine, skin, and larynx in addition to the colon. Diagnosis of classical HNPCC is based on the Amsterdam criteria: 3 or more relatives affected by colorectal cancer, one a first degree relative of the other two; 2 or more generation affected; 1 or more colorectal cancers presenting before 50 years of age; exclusion of hereditary polyposis syndromes. MSH6 mutations appear to be associated with atypical HNPCC and in particular with development of endometrial carcinoma or atypical endometrial hyperplasia, the presumed precursor of endometrial cancer. Defects in MSH6 are also found in familial colorectal cancers (suspected or incomplete HNPCC) that do not fulfill the Amsterdam criteria for HNPCC.

Defects in MSH6 are a cause of susceptibility to endometrial cancer (ENDMC) [MIM:608089].

序列相似性 Belongs to the DNA mismatch repair mutS family.

Contains 1 PWWP domain.

翻译后修饰 The N-terminus is blocked.

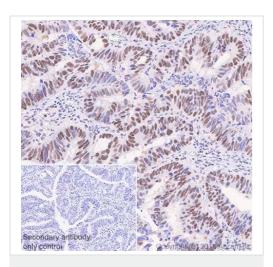
Phosphorylated upon DNA damage, probably by ATM or ATR.

Phosphorylated by PRKCZ, which may prevent MutS alpha degradation by the ubiquitin-

proteasome pathway.

细胞定位 Nucleus.

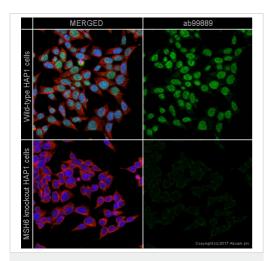
图片



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MSH6 antibody [SP93] (ab99889)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human colon carcinoma tissue sections labeling MSH6 with ab99889 at 1/100 dilution (1.0 µg/ml). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10mins. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Hematoxylin was used as a counterstain. Nuclear staining on human colon carcinoma, performed on a Leica Biosystems BOND™ RX instrument.

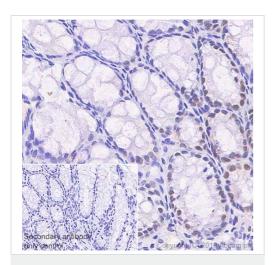
The section was incubated with ab99889 for 30 mins at room temperature.



Immunocytochemistry/ Immunofluorescence - Anti-MSH6 antibody [SP93] (ab99889)

ab99889 staining MSH6 in wild-type HAP1 cells (top panel) and MSH6 knockout HAP1 cells (bottom panel). The cells were fixed with 100% methanol (5min), 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 ab99889 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 lgG (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 paraffinembedded sections) - Anti-MSH6 antibody [SP93] (ab99889)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Mouse colon

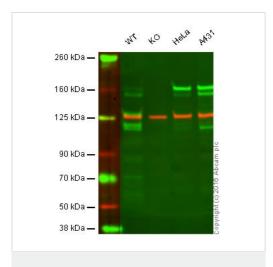
tissue sections labeling MSH6 with ab99889 at 1/100 dilution (1.0 µg/ml). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10mins. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Hematoxylin was used as a counterstain. Sporadically nuclear staining on mouse colon, performed on a Leica Biosystems BOND™ RX instrument.

The section was incubated with ab99889 for 30 mins at room temperature.



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MSH6 antibody [SP93] (ab99889)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human colon tissue sections labeling MSH6 with ab99889 at 1/100 dilution (1.0 μg/ml). Heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0, epitope retrieval solution 2) for 10mins. Rabbit specific IHC polymer detection kit HRP/DAB (ab209101) was used as the secondary antibody. Hematoxylin was used as a counterstain. Sporadically nuclear staining on human colon, performed on a Leica Biosystems BONDTM RX instrument. The section was incubated with ab99889 for 30 mins at room temperature.



Western blot - Anti-MSH6 antibody [SP93] (ab99889)

Lane 1: Wild-type HAP1 cell lysate (20 µg)

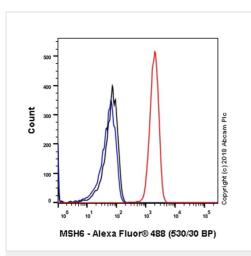
Lane 2: MSH6 knockout HAP1 cell lysate (20 µg)

Lane 3: HeLa cell lysate (20 µg)

Lane 4: A431 cell lysate (20 µg)

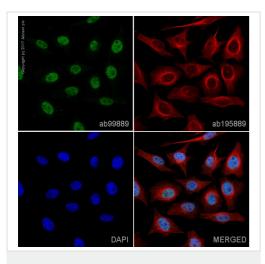
Lanes 1 - 4: Merged signal (red and green). Green - ab99889 observed at 160 kDa. Red - loading control, **ab18058**, observed at 124 kDa.

ab99889 was shown to specifically react with MSH6 in wild-type HAP1 cells along with additional cross reactive bands. No band was observed in MSH6 knockout samples. Wild-type and MSH6 knockout samples were subjected to SDS-PAGE. ab99889 and ab18058 (loading control to Vinculin) were diluted at 1 µg/ml and 1/10 000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed ab216776 secondary antibodies at 1/10 000 dilution for 1 h at room temperature before imaging.



Flow Cytometry (Intracellular) - Anti-MSH6 antibody [SP93] (ab99889)

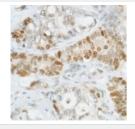
Flow cytometry analysis of Hela (human cervix adenocarcinoma) labeling MSH6 with purified ab99889 at 1/20 dilution (5.05 μ g/ml) (red). Cells were fixed with 4% paraformaldehyde and permeabilised with 90% methanol. Goat anti rabbit lgG (Alexa Fluor[®] 488, **ab150077**) at 1/2000 dilution was used as a secondary antibody. Isotype control - Rabbit monoclonal lgG (**ab172730**) (Black). Unlableled control - Unlabelled cells (blue).



Immunocytochemistry/ Immunofluorescence - Anti-MSH6 antibody [SP93] (ab99889)

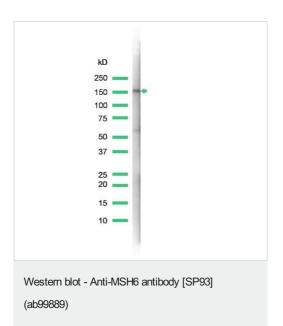
ab99889 staining MSH6 in HeLa cells. The cells were fixed with 100% methanol (5min), permeabilized with 0.1% Triton X-100 for 5 minutes and then blocked with 1% BSA/10% normal goat serum/0.3M glycine in 0.1% PBS-Tween for 1h. The cells were then incubated with ab99889 at 1/250 dilution and **ab195889** 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).



Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-MSH6 antibody [SP93] (ab99889)

Staining of MSH6 in a formalin fixed, paraffin embedded Human rectal carcinoma tissue using ab99889 at a dilution of 1/100.



Anti-MSH6 antibody [SP93] (ab99889) at 1/100 dilution + Lysate prepared from A431 cells

Observed band size: 170 kDa



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