abcam

Product datasheet

Anti-MTAP antibody [EPR6893] ab126770

KO VALIDATED Recombinant RabMAb

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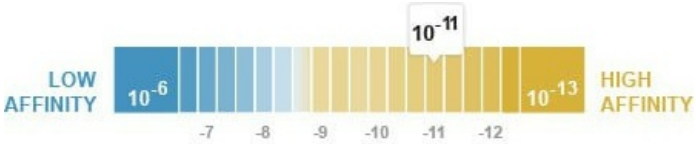
Overview

Product name	Anti-MTAP antibody [EPR6893]
Description	Rabbit monoclonal [EPR6893] to MTAP
Host species	Rabbit
Specificity	The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat.
Tested applications	**Suitable for:** Flow Cyt (Intra), WB, IP, IHC-P, ICC/IF
Species reactivity	**Reacts with:** Mouse, Rat, Human
Immunogen	Synthetic peptide within Human MTAP aa 200-300. The exact sequence is proprietary. Database link: **Q13126**
Positive control	WB: HeLa, 293T, HT29, C6, RAW 264.7, and NIH 3T3 cell lysates. ICC/IF: HeLa cells. Flow Cyt (intra): HeLa cells. IHC-P: Human kidney, mouse kidney, and human lung carcinoma tissue.
General notes	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**. Our RabMAb® technology is a patented hybridoma-based technology for making rabbit monoclonal antibodies. For details on our patents, please refer to **RabMAb® patents**.

Properties

Form	Liquid
Storage instructions	Shipped at 4°C. Store at +4°C short term (1-2 weeks). Upon delivery aliquot. Store at -20°C long term. Avoid freeze / thaw cycle.
Dissociation constant (K_D)	$K_D = 2.60 \times 10^{-11}$ M



Learn more about K_D

Storage buffer	pH: 7.20 Preservative: 0.01% Sodium azide Constituents: 59% PBS, 40% Glycerol (glycerin, glycerine), 0.5% BSA
Purity	Protein A purified
Clonality	Monoclonal
Clone number	EPR6893
Isotype	IgG

Applications

The Abpromise guarantee Our **Abpromise guarantee** covers the use of ab126770 in the following tested applications.

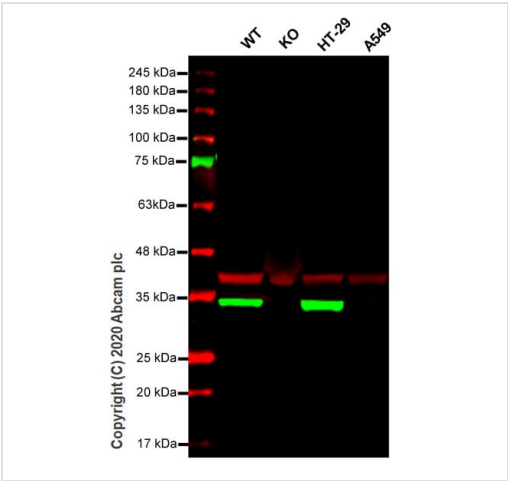
The application notes include recommended starting dilutions; optimal dilutions/concentrations should be determined by the end user.

Application	Abreviews	Notes
Flow Cyt (Intra)		1/90. **ab172730** - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody. **For unpurified use at 1/100 - 1/500.**
WB		1/1000 - 1/10000. Detects a band of approximately 29 kDa (predicted molecular weight: 31 kDa).
IP		1/10 - 1/100.
IHC-P	★★★☆☆ (1)	1/1000. Perform heat mediated antigen retrieval before commencing with IHC staining protocol. The mouse and rat recommendation is based on the WB results. We do not guarantee IHC-P for mouse and rat **For unpurified use at 1:50 - 1:100.**
ICC/IF		1/50 - 1/250.

Target

Function	Plays a major role in polyamine metabolism and is important for the salvage of both adenine and methionine.
Tissue specificity	Ubiquitously expressed.
Sequence similarities	Belongs to the PNP/MTAP phosphorylase family.
Cellular localization	Cytoplasm.

Images



Western blot - Anti-MTAP antibody [EPR6893] (ab126770)

All lanes : Anti-MTAP antibody [EPR6893] (ab126770) at 1/1000 dilution

Lane 1 : Wild-type HeLa cell lysate
Lane 2 : MTAP knockout HeLa cell lysate
Lane 3 : HT-29 cell lysate
Lane 4 : A549 cell lysate

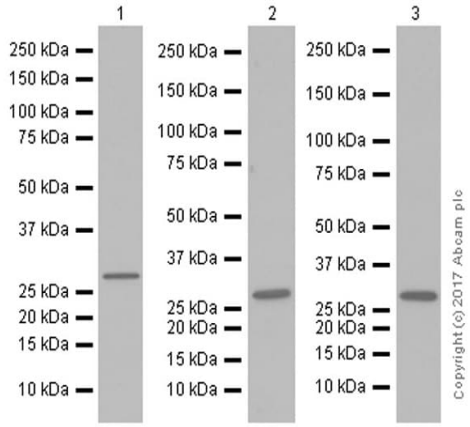
Lysates/proteins at 20 µg per lane.

Performed under reducing conditions.

Predicted band size: 31 kDa
Observed band size: 32 kDa

anes 1- 4: Merged signal (red and green). Green - ab126770 observed at 32 kDa. Red - Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) observed at 37 kDa.

ab126770 Anti-MTAP antibody [EPR6893] was shown to specifically react with MTAP in wild-type HeLa cells. Loss of signal was observed when knockout cell line **ab265272** (knockout cell lysate **ab257194**) was used. Wild-type and MTAP knockout samples were subjected to SDS-PAGE. ab126770 and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4°C at 1 in 1000 dilution and 1 in 20000 dilution respectively. 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 in 10000 dilution for 1 hour at room temperature before imaging.



Western blot - Anti-MTAP antibody [EPR6893] (ab126770)

All lanes : Anti-MTAP antibody [EPR6893] (ab126770) at 1/10000 dilution (purified)

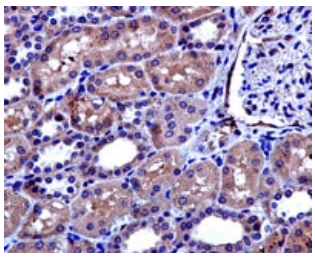
Lane 1 : NIH/3T3 (Mouse embryonic fibroblast) whole cell lysates
Lane 2 : C6 (Rat glial tumor glial cell) whole cell lysates
Lane 3 : Mouse kidney lysates

Lysates/proteins at 15 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 31 kDa

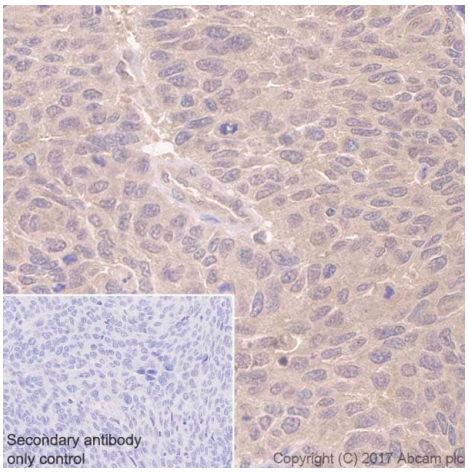
Blocking and diluting buffer : 5% NFDM/TBST



Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MTAP antibody [EPR6893] (ab126770)

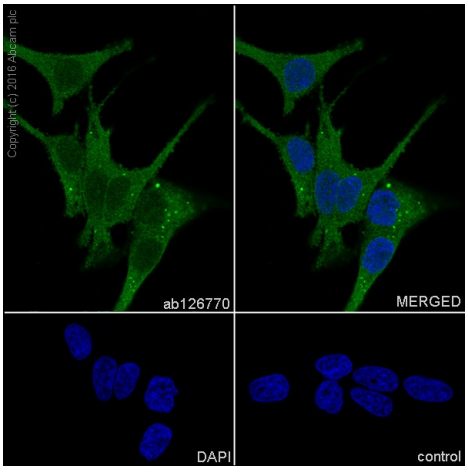
Formalin-fixed, paraffin-embedded human kidney tissue stained for MTAP with unpurified ab126770 (1/50 dilution) in immunohistochemical analysis.

Heat mediated antigen retrieval was performed before commencing with IHC staining protocol.



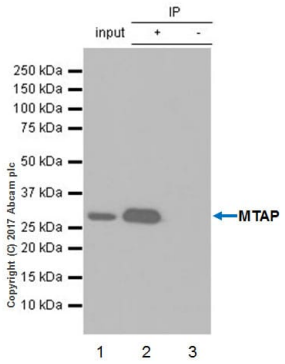
Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) - Anti-MTAP antibody [EPR6893] (ab126770)

Immunohistochemistry (Formalin/PFA-fixed paraffin-embedded sections) analysis of Human lung carcinoma tissue sections labeling MTAP with Purified ab126770 at 1:1000 dilution (0.89 µg/ml). Heat mediated antigen retrieval was performed using **ab93684** (Tris/EDTA buffer, pH 9.0). Tissue was counterstained with Hematoxylin. ImmunoHistoProbe one step HRP Polymer (ready to use) secondary antibody was used at 1:0 dilution. PBS instead of the primary antibody was used as the negative control.



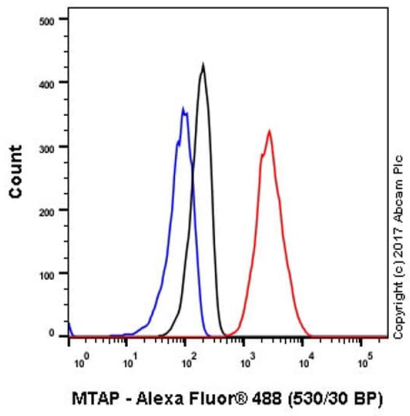
Immunocytochemistry/ Immunofluorescence - Anti-MTAP antibody [EPR6893] (ab126770)

Immunocytochemistry/ Immunofluorescence analysis of HeLa (Human epithelial cell line from cervix adenocarcinoma) labeling MTAP with Purified ab126770 at 1/250 dilution. Cells were fixed with 4% PFA and permeabilized with 0.1% tritonX-100. **ab150077** Goat anti rabbit IgG (Alexa Fluor® 488) at 1/1000 was used as the secondary antibody. Nuclei were counterstained with DAPI. PBS was used instead of the primary antibody as the negative control.



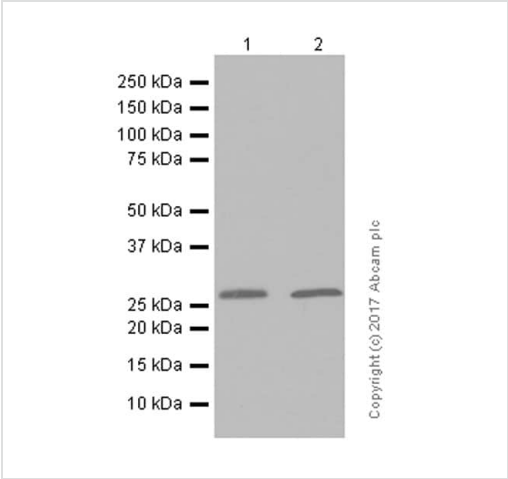
Immunoprecipitation - Anti-MTAP antibody [EPR6893] (ab126770)

ab126770 (purified) at 1:50 dilution (2µg) immunoprecipitating MTAP in HT-29 whole cell lysate.
Lane 1 (input): HT-29 (Human colorectal adenocarcinoma epithelial cell) whole cell lysate 10ug
Lane 2 (+): ab126770 & HT-29 whole cell lysate
Lane 3 (-): Rabbit monoclonal IgG (**ab172730**) instead of ab126770 in HT-29 whole cell lysate
For western blotting, VeriBlot for IP Detection Reagent (HRP) (**ab131366**) was used for detection at 1:1000 dilution.
Blocking and diluting buffer: 5% NFDM/TBST.



Flow Cytometry (Intracellular) - Anti-MTAP antibody [EPR6893] (ab126770)

Intracellular Flow Cytometry analysis of HeLa (Human cervix adenocarcinoma epithelial cell) cells labeling MTAP with purified ab126770 at 1/90 dilution (10 µg/ml) (red). Cells were fixed with 4% Paraformaldehyde. A Goat anti rabbit IgG (Alexa Fluor® 488) secondary antibody was used at 1/2000 dilution. Isotype control - Rabbit monoclonal IgG (Black). Unlabeled control - Cell without incubation with primary antibody and secondary antibody (Blue).



Western blot - Anti-MTAP antibody [EPR6893] (ab126770)

All lanes : Anti-MTAP antibody [EPR6893] (ab126770) at 1/2000 dilution (purified)

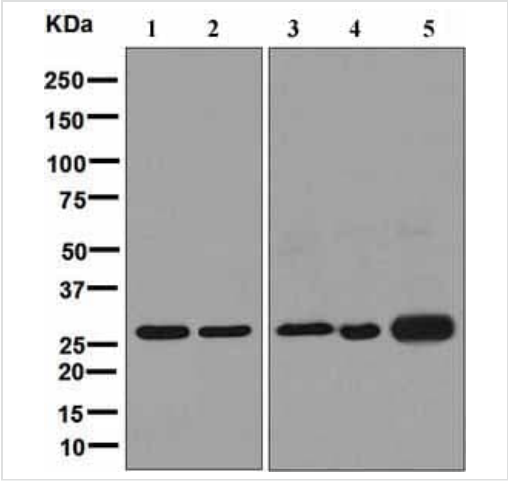
Lane 1 : HT-29 (Human colorectal adenocarcinoma epithelial cell) whole cell lysates
Lane 2 : HEK-293 (Human embryonic kidney epithelial cell) whole cell lysates

Lysates/proteins at 20 µg per lane.

Secondary
All lanes : Goat Anti-Rabbit IgG H&L (HRP) (**ab97051**) at 1/20000 dilution

Predicted band size: 31 kDa

Blocking and diluting buffer : 5% NFDM/TBST



Western blot - Anti-MTAP antibody [EPR6893] (ab126770)

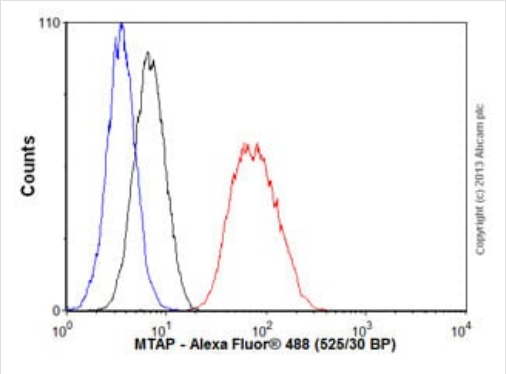
All lanes : Anti-MTAP antibody [EPR6893] (ab126770) at 1/1000 dilution (unpurified)

Lane 1 : 293T cell lysates
Lane 2 : HT29 cell lysates
Lane 3 : C6 cell lysates
Lane 4 : RAW 264.7 cell lysates
Lane 5 : NIH 3T3 cell lysates

Lysates/proteins at 10 µg per lane.

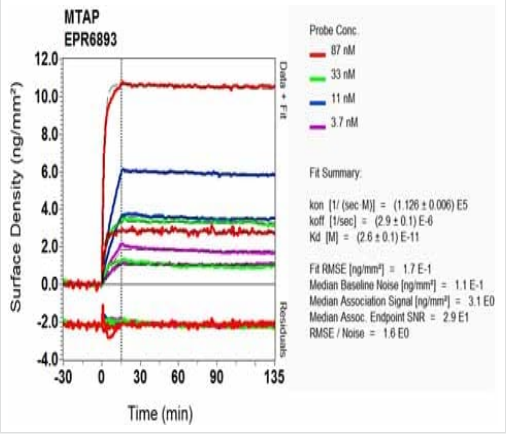
Secondary
All lanes : Goat anti-Rabbit HRP at 1/2000 dilution

Predicted band size: 31 kDa



Flow Cytometry (Intracellular) - Anti-MTAP antibody [EPR6893] (ab126770)

Overlay histogram showing HeLa cells stained with unpurified ab126770 (red line). The cells were fixed with 80% methanol (5 min) and then permeabilized with 0.1% PBS-Tween for 20 min. The cells were then incubated in 1x PBS / 10% normal goat serum / 0.3M glycine to block non-specific protein-protein interactions followed by the antibody (ab126770, 1/100 dilution) for 30 min at 22°C. The secondary antibody used was Alexa Fluor® 488 goat anti-rabbit IgG (H&L) (**ab150077**) at 1/2000 dilution for 30 min at 22°C. Isotype control antibody (black line) was rabbit IgG (monoclonal) (1µg/1x10^6 cells) used under the same conditions. Unlabelled sample (blue line) was also used as a control. Acquisition of >5,000 events were collected using a 20mW Argon ion laser (488nm) and 525/30 bandpass filter.

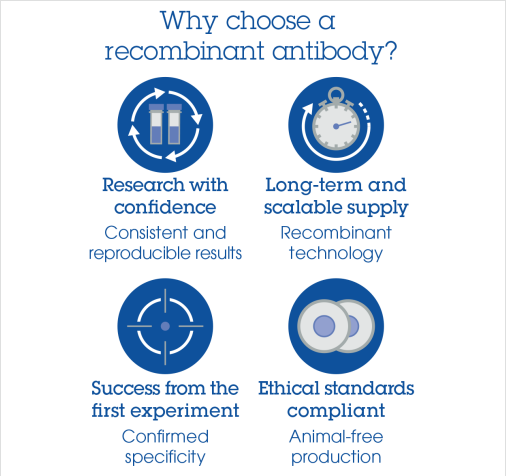


OI-RD Scanning - Anti-MTAP antibody [EPR6893] (ab126770)

Equilibrium disassociation constant (K_D)

Learn more about K_D

Click here to learn more about K_D



Anti-MTAP antibody [EPR6893] (ab126770)

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