## abcam

## Product datasheet

## Anti－MTAP antibody［EPR6893］ab126770 <br> 

## 青青帾 1 Abreviews 8 References 画像数 12

## 製品の概要

| 製品名 | Anti－MTAP antibody［EPR6893］ |
| :---: | :---: |
| 製品の詳細 | Rabbit monoclonal［EPR6893］to MTAP |
| 由来種 | Rabbit |
| 特異性 | The mouse and rat recommendation is based on the WB results．We do not guarantee IHC－P for mouse and rat． |
| アプリケーション | 適用あり：Flow Cyt（Intra），WB，IP，IHC－P，ICC／IF |
| 種交差性 | 交差種：Mouse，Rat，Human |
| 免疫原 | Synthetic peptide within Human MTAP aa 200－300．The exact sequence is proprietary． Database link：Q13126 |
| ポジティブ・コントロール | 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． |
| 特記事項 | 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 ${ }^{\circledR}$ technology is a patented hybridoma－based technology for making rabbit monoclonal antibodies．For details on our patents，please refer to $\mathbf{R a b M A b}^{\circledR}$ patents． |

製品の特性

## 製品の状態

保存方法

## 解離定数（ $K_{D}$ 値）

Liquid
Shipped at $4^{\circ} \mathrm{C}$ ．Store at $+4^{\circ} \mathrm{C}$ short term（ $1-2$ weeks）．Upon delivery aliquot．Store at $-20^{\circ} \mathrm{C}$ long term．Avoid freeze／thaw cycle．
$K_{D}=2.60 \times 10^{-11} \mathrm{M}$


## Learn more about $K_{\underline{D}}$

| バッファー | pH： 7.20 |  |
| :---: | :---: | :---: |
|  | Preservative：0．01\％Sodium azide |  |
|  | Constituents：59\％PBS，40\％Glycerol（glycerin，glycerine），0．5\％BSA |  |
| 精製度 | Protein A purified |  |
| ポリノモノ | モノクローナル |  |
| クローン名 | EPR6893 |  |
| アイソタイプ | $\lg \mathrm{G}$ |  |
| アプリケーション |  |  |
| The Abpromise guarantee | Abpromise保証は，次のテスト済みアプリナーションにおけるab126770の使用に適用されます |  |
| アプリケーションノートには，推奨の開始希釈率がありますが，適切な希釈率につきましてはご検討ください。 |  |  |
| アプリケーション | Abreviews | 特記事項 |
| Flow Cyt（Intra） |  | 1／90． <br> ab172730－Rabbit monoclonal lgG，is suitable for use as an isotype control with this antibody． <br> 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 | 为为为能（1） | 1／1000．Perform heat mediated antigen retrieval before commencing with IHC staining protocol． <br> The mouse and rat recommendation is based on the WB results． We do not guarantee IHC－P for mouse and rat <br> For unpurified use at 1：50－1：100． |
| ICC／IF |  | 1／50－1／250． |

## ターゲット情報

| 機能 | Plays a major role in polyamine metabolism and is important for the salvage of both adenine and <br> methionine． |
| :--- | :--- |
| 組織特異性 | Ubiquitously expressed． |
| 配列類似性 | Belongs to the PNP／MTAP phosphorylase family． |
| 細胞内局在 | Cytoplasm． |

画像


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 $20 \mu \mathrm{~g}$ 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 AntiGAPDH antibody [6C5] - Loading Control (ab8245) were incubated overnight at $4^{\circ} \mathrm{C}$ at 1 in 1000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H\&L (IRDye ${ }^{\circledR}$ 800CW) preadsorbed (ab216773) and Goat anti-Mouse $\operatorname{lgG} H \& L\left(I R D y e^{\circledR} 680 R D\right)$ 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 $15 \mu \mathrm{~g}$ per lane.

## Secondary

All lanes: Goat Anti-Rabbit lgG H\&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 31 kDa

## Blocking and diluting buffer : $5 \%$ NFDM/TBST

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) analysis of Human lung carcinoma tissue sections labeling MTAP with Purified ab126770 at 1:1000 dilution (0.89 $\mu \mathrm{g} / \mathrm{ml})$. 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 - AntiMTAP 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 lgG (Alexa Fluor ${ }^{\circledR}$ 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 $(2 \mu \mathrm{~g})$ 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 lgG (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.

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


Westem 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 $20 \mu \mathrm{~g}$ per lane.

## Secondary

All lanes: Goat Anti-Rabbit lgG H\&L (HRP) (ab97051) at 1/20000 dilution

Predicted band size: 31 kDa

Blocking and diluting buffer : 5\% NFDM/TBST


Westem 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 $10 \mu \mathrm{~g}$ 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 / 0.3 M glycine to block non-specific protein-protein interactions followed by the antibody (ab126770, 1/100 dilution) for 30 min at $22^{\circ} \mathrm{C}$. The secondary antibody used was Alexa Fluor ${ }^{\circledR} 488$ goat anti-rabbit lgG (H\&L) (ab150077) at 1/2000 dilution for 30 min at $22^{\circ} \mathrm{C}$. Isotype control antibody (black line) was rabbit lgG (monoclonal) $\left(1 \mu \mathrm{~g} / 1 \times 10^{6}\right.$ 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 20 mW Argon ion laser (488nm) and 525/30 bandpass filter.

Equilibrium disassociation constant ( $\mathrm{K}_{\mathrm{D}}$ ) Learn more about $K_{D}$

## Click here to learn more about $K_{\underline{D}}$

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