# abcam

## Product datasheet

# Anti-WTAP antibody [EPR18744] ab195380





重组 RabMAb

★★★★★ 6 Abreviews 39 References 10 图像

概述

产品名称 Anti-WTAP抗体[EPR18744]

描述 兔单克隆抗体[EPR18744] to WTAP

宿主 Rabbit

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

种属反应性 与反应: Human

预测可用于: Pig, Non human primates ◆ 不与反应: Mouse

免疫原 Recombinant fragment. This information is proprietary to Abcam and/or its suppliers.

阳性对照 WB: Jurkat, K562, HeLa and HepG2 whole cell lysates; human kidney, thymus and lung lysates.

IHC-P: Human endometrium and liver cancer tissues. ICC/IF: Jurkat and HeLa cells. Flow Cyt

(intra): Jurkat cells. IP: Jurkat whole cell lysate.

常规说明 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**.

性能

形式

存放说明 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.

存储溶液 pH: 7.2

Preservative: 0.01% Sodium azide

Constituents: PBS, 40% Glycerol (glycerin, glycerine), 0.05% BSA

纯度 Protein A purified

克隆 单克隆

**克隆编号** EPR18744

**同种型** IgG

#### 应用

### The Abpromise guarantee Abpromise™承诺保证使用ab195380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
Flow Cyt (Intra)		1/600.
ICC/IF		1/500.
IP		1/40.
IHC-P	<b>★★★★★</b> (3)	1/100. Perform heat mediated antigen retrieval with Tris/EDTA buffer pH 9.0 before commencing with IHC staining protocol.
WB	**** <u>(2)</u>	1/1000. Detects a band of approximately 50 kDa (predicted molecular weight: 44 kDa). Milk recommended as blocking agent.

靶标

功能 Regulates G2/M cell-cycle transition by binding to the 3' UTR of CCNA2, which enhances its

stability. Impairs WT1 DNA-binding ability and inhibits expression of WT1 target genes. May be

involved in mRNA splicing regulation.

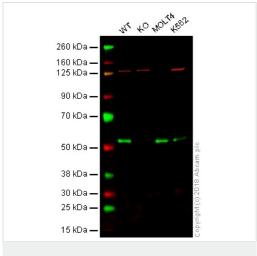
组织**特异性** Ubiquitously expressed.

序列相似性 Belongs to the fl(2)d family.

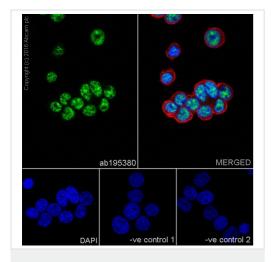
翻译后修饰 Phosphorylated upon DNA damage, probably by ATM or ATR.

细胞定位 Nucleus > nucleolus.

#### 图片



Western blot - Anti-WTAP antibody [EPR18744] (ab195380)



Immunocytochemistry/ Immunofluorescence - Anti-WTAP antibody [EPR18744] (ab195380)

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

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

Lane 3: MOLT-4 whole cell lysate (20 µg)

Lane 4: K562 whole cell lysate (20 µg)

**Lanes 1 - 4:** Merged signal (red and green). Green - ab195380 observed at 50 kDa. Red - loading control, <u>ab18058</u>, observed at 130 kDa.

ab195380 was shown to specifically react with WTAP in wild-type HAP1 cells as signal was lost in WTAP knockout cells. Wild-type and WTAP knockout samples were subjected to SDS-PAGE.

Ab195380 and ab18058 (Mouse anti-Vinculin loading control) were incubated overnight at 4°C at 1/1000 dilution and 1/20000 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 1/10000 dilution for 1 hour at room temperature before imaging.

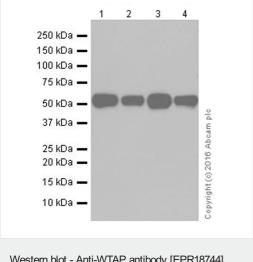
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized Jurkat (Human T cell leukemia cell line from peripheral blood) cells labeling WTAP with ab195380 at 1/500 dilution, followed by Goat Anti-Rabbit IgG (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on Jurkat cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (<u>ab7291</u>) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) (<u>ab150120</u>) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab195380 at 1/500 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) (ab150120) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (<u>ab7291</u>) at 1/1000 dilution, followed by Goat Anti-Rabbit lgG H&L (Alexa Fluor<sup>®</sup> 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution.



Western blot - Anti-WTAP antibody [EPR18744] (ab195380)

**All lanes :** Anti-WTAP antibody [EPR18744] (ab195380) at 1/1000 dilution

**Lane 1 :** Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate

**Lane 2**: K562 (Human chronic myelogenous leukemia cell line from bone marrow) whole cell lysate

**Lane 3**: HeLa (Human epithelial cell line from cervix adenocarcinoma) whole cell lysate

**Lane 4 :** HepG2 (Human liver hepatocellular carcinoma cell line) whole cell lysate

Lysates/proteins at 20 µg per lane.

#### **Secondary**

**All lanes :** Goat Anti-Rabbit lgG H&L (HRP) (<u>ab97051</u>) at 1/100000 dilution

Developed using the ECL technique.

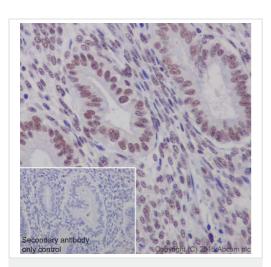
**Predicted band size:** 44 kDa **Observed band size:** 50 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

The observed molecular weight is consistent with what has been

described in the literature (PMID: 17088532).

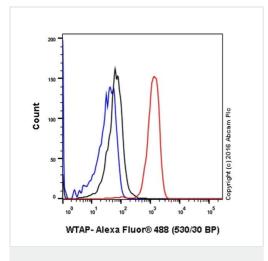


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-WTAP antibody
[EPR18744] (ab195380)

Immunohistochemical analysis of paraffin-embedded human endometrium tissue labeling WTAP with ab195380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on human endometrium is observed. Counter stained with Hematoxylin.

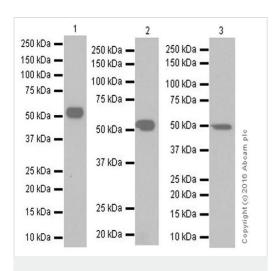
Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

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



Flow Cytometry (Intracellular) - Anti-WTAP antibody [EPR18744] (ab195380)

Intracellular flow cytometric analysis of 4% paraformaldehyde-fixed Jurkat (Human T cell leukemia cell line from peripheral blood) cells labeling WTAP with ab195380 at 1/600 dilution (red) compared with a rabbit monoclonal IgG isotype control (ab172730; black) and an unlabelled control (cells without incubation with primary antibody and secondary antibody; blue). Goat Anti-Rabbit IgG (Alexa Fluorr® 488) at 1/2000 dilution was used as the secondary antibody.



Western blot - Anti-WTAP antibody [EPR18744] (ab195380)

**All lanes :** Anti-WTAP antibody [EPR18744] (ab195380) at 1/1000 dilution

Lane 1 : Human kidney lysate
Lane 2 : Human thymus lysate
Lane 3 : Human lung lysate

Lysates/proteins at 10 µg per lane.

#### Secondary

**All lanes :** Goat Anti-Rabbit IgG Peroxidase Conjugate, specific to the non-reduced form of IgG at 1/100000 dilution

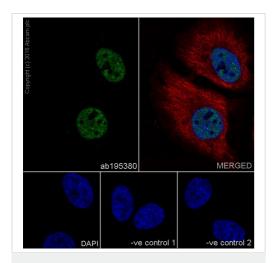
Developed using the ECL technique.

**Predicted band size:** 44 kDa **Observed band size:** 50 kDa

Exposure time: 3 minutes

Blocking/Dilution buffer: 5% NFDM/TBST.

The observed molecular weight is consistent with what has been described in the literature (PMID: 17088532).



Immunocytochemistry/ Immunofluorescence - Anti-WTAP antibody [EPR18744] (ab195380)

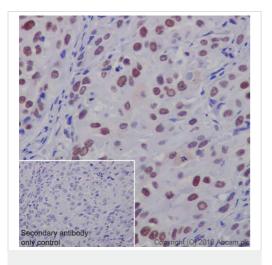
Immunofluorescent analysis of 4% paraformaldehyde-fixed, 0.1% Triton X-100 permeabilized HeLa (Human epithelial cell line from cervix adenocarcinoma) cells labeling WTAP with ab195380 at 1/500 dilution, followed by Goat Anti-Rabbit lgG (Alexa Fluor<sup>®</sup> 488) (ab150077) secondary antibody at 1/1000 dilution (green). Confocal image showing nuclear staining on HeLa cell line. The nuclear counter stain is DAPI (blue).

Tubulin is detected with Anti-alpha Tubulin mouse MAb (<u>ab7291</u>) at 1/1000 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor<sup>®</sup> 594) (<u>ab150120</u>) secondary antibody at 1/1000 dilution (red).

The negative controls are as follows:

-ve control 1: ab195380 at 1/500 dilution, followed by Goat Anti-Mouse IgG H&L (Alexa Fluor® 594) (ab150120) secondary antibody at 1/1000 dilution.

-ve control 2: Anti-alpha Tubulin mouse MAb (<u>ab7291</u>) at 1/1000 dilution, followed by Goat Anti-Rabbit lgG (Alexa Fluor<sup>®</sup> 488) (<u>ab150077</u>) secondary antibody at 1/1000 dilution.

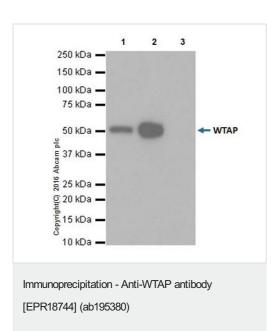


Immunohistochemistry (Formalin/PFA-fixed paraffinembedded sections) - Anti-WTAP antibody
[EPR18744] (ab195380)

Immunohistochemical analysis of paraffin-embedded human liver cancer tissue labeling WTAP with ab195380 at 1/100 dilution, followed by Goat Anti-Rabbit IgG H&L (HRP) (ab97051) at 1/500 dilution. Nuclear staining on human liver cancer is observed. Counter stained with Hematoxylin.

Secondary antibody only control: Used PBS instead of primary antibody, secondary antibody is Goat Anti-Rabbit lgG H&L (HRP) (ab97051) at 1/500 dilution.

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



WTAP was immunoprecipitated from 0.35 mg of Jurkat (Human T cell leukemia cell line from peripheral blood) whole cell lysate with ab195380 at 1/40 dilution. Western blot was performed from the immunoprecipitate using ab195380 at 1/1000 dilution. VeriBlot for IP Detection Reagent (HRP) (ab131366), was used for detection at 1/10000 dilution.

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

Lane 2: ab195380 IP in Jurkat whole cell lysate.

Lane 3: Rabbit monoclonal  $\lg G$  ( $\underline{ab172730}$ ) instead of ab195380 in Jurkat whole cell lysate.

Blocking and dilution buffer and concentration: 5% NFDM/TBST.

Exposure time: 30 seconds.



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