# **Anti-WTAP Antibody**

### **HA500525**



Product Type: Rabbit polyclonal IgG, primary antibodies

Species reactivity: Human, Mouse, Rat
Applications: WB, IHC-P, IF-Cell

Molecular Wt: Predicted band size: 44 kDa

**Description:** Pre-mRNA-splicing regulator WTAP is a protein that in humans is encoded by the WTAP

gene. The Wilms tumor suppressor gene WT1 appears to play a role in both transcriptional and posttranscriptional regulation of certain cellular genes. This gene encodes a WT1-associating protein, which is a ubiquitously expressed nuclear protein. Like WT1 protein, this protein is localized throughout the nucleoplasm as well as in speckles and partially colocalizes with splicing factors. Alternative splicing of this gene results in three transcript variants, two of which encode the same isoform. WTAP (gene) has been shown to interact

with WT1.

**Immunogen:** Recombinant protein within human WTAP aa 1-300 / 396.

Positive control: K562 cell lysate, Jurkat cell lysate, Hela cell lysate, HepG2 cell lysate, 293T cell lysate,

NIH/3T3 cell lysate, PC-12 cell lysate, human breast carcinoma tissue, human colon tissue,

mouse small intestine tissue, rat small intestine tissue, NIH/3T3.

Subcellular location: Nucleus speckle, Nucleus, nucleoplasm, Cytoplasm.

Database links: SwissProt: Q15007 Human | Q9ER69 Mouse

Entrez Gene: 499020 Rat

**Recommended Dilutions:** 

WB 1:1,000 IHC-P 1:2,000 IF-Cell 1:200

Storage Buffer: PBS (pH7.4), 0.1% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

**Storage Instruction:** Store at  $+4^{\circ}$ C after thawing. Aliquot store at  $-20^{\circ}$ C. Avoid repeated freeze / thaw cycles.

Purity: Immunogen affinity purified.

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#### **Images**

 **Fig1:** Western blot analysis of WTAP on different lysates with Rabbit anti-WTAP antibody (HA500525) at 1/1,000 dilution.

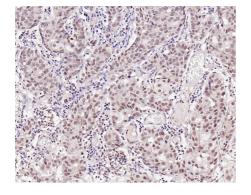
Lane 1: K562 cell lysate (12.5 µg/Lane) Lane 2: Jurkat cell lysate (10 µg/Lane) Lane 3: Hela cell lysate (10 µg/Lane) Lane 4: HepG2 cell lysate (10 µg/Lane) Lane 5: 293T cell lysate (10 µg/Lane) Lane 6: NIH/3T3 cell lysate (10 µg/Lane) Lane 7: PC-12 cell lysate (10 µg/Lane)

Predicted band size: 44 kDa Observed band size: 55 kDa

Exposure time: 30 seconds;

4-20% SDS-PAGE gel.

Proteins were transferred to a PVDF membrane and blocked with 5% NFDM/TBST for 1 hour at room temperature. The primary antibody (HA500525) at 1/1,000 dilution was used in 5% NFDM/TBST at room temperature for 2 hours. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:200,000 dilution was used for 1 hour at room temperature.

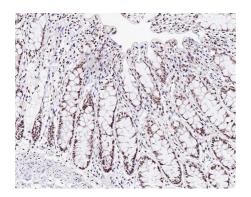


**Fig2:** Immunohistochemical analysis of paraffin-embedded human breast carcinoma tissue with Rabbit anti-WTAP antibody (HA500525) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA500525) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

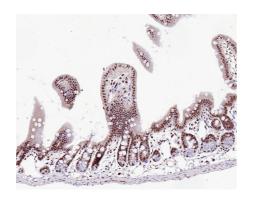
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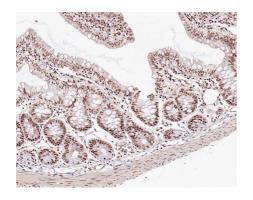
**Fig3:** Immunohistochemical analysis of paraffin-embedded human colon tissue with Rabbit anti-WTAP antibody (HA500525) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH $_2$ O and PBS, and then probed with the primary antibody (HA500525) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig4:** Immunohistochemical analysis of paraffin-embedded mouse small intestine tissue with Rabbit anti-WTAP antibody (HA500525) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA500525) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.



**Fig5:** Immunohistochemical analysis of paraffin-embedded rat small intestine tissue with Rabbit anti-WTAP antibody (HA500525) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with sodium citrate buffer (pH 6.0) for 2 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH<sub>2</sub>O and PBS, and then probed with the primary antibody (HA500525) at 1/2,000 dilution for 1 hour at room temperature. The detection was performed using an HRP conjugated compact polymer system. DAB was used as the chromogen. Tissues were counterstained with hematoxylin and mounted with DPX.

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Fig6: Immunocytochemistry analysis of NIH/3T3 cells labeling WTAP with Rabbit anti-WTAP antibody (HA500525) at  $10\mu g/mL$  dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37  $^{\circ}$ C, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-WTAP antibody (HA500525) at 10 $\mu$ g/mL dilution in 2% negative goat serum overnight at 4  $^{\circ}$ C. Goat Anti-Rabbit IgG H&L (iFluor 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

Beta tubulin (M1305-2, red) was stained at 1/100 dilution overnight at  $+4^{\circ}$ C. Goat Anti-Mouse IgG H&L (iFluor  $^{\dagger}$  594, HA1126) was used as the secondary antibody at 1/1,000 dilution.

Note: All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE".

#### **Background References**

- 1. Li ZX et al. WTAP-mediated m6A modification of IncRNA DIAPH1-AS1 enhances its stability to facilitate nasopharyngeal carcinoma growth and metastasis. Cell Death Differ. 2022 Jun
- 2. Li G et al. WTAP-mediated m6A modification of IncRNA NORAD promotes intervertebral disc degeneration. Nat Commun. 2022 Mar