

Anti-PUMA Antibody [SR42-09] - BSA and Azide free

HA750038



Product Type:	Recombinant Rabbit monoclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IF-Tissue, IHC-P, FC
Molecular Wt:	Predicted band size: 21 kDa
Clone number:	SR42-09

Description: The p53 upregulated modulator of apoptosis (PUMA) also known as Bcl-2-binding component 3 (BBC3), is a pro-apoptotic protein, member of the Bcl-2 protein family. In humans, the Bcl-2-binding component 3 protein is encoded by the BBC3 gene. The expression of PUMA is regulated by the tumor suppressor p53. PUMA is involved in p53-dependent and -independent apoptosis induced by a variety of signals, and is regulated by transcription factors, not by post-translational modifications. After activation, PUMA interacts with antiapoptotic Bcl-2 family members, thus freeing Bax and/or Bak which are then able to signal apoptosis to the mitochondria. Following mitochondrial dysfunction, the caspase cascade is activated ultimately leading to cell death.

Immunogen: Synthetic peptide within Human PUMA aa 153-193 / 193.

Positive control: HeLa cell lysate, HepG2 cell lysate, HDLM-2 cell lysate, NIH/3T3 cell lysate, Rat stomach tissue lysate, Rat liver tissue lysate, Hela, SKOV-3, human stomach cancer tissue, human small intestine tissue, mouse small intestine tissue, rat small intestine tissue, rat stomach tissue.

Subcellular location: Mitochondrion.

Database links: SwissProt: Q9BXH1 Human | Q99ML1 Mouse | Q80ZG6 Rat

Recommended Dilutions:

WB	1:500-1:2,000
IF-Cell	1:50
IF-Tissue	1:200-1:500
IHC-P	1:2,000-1:30,000
FC	1:50

Storage Buffer: 1*PBS (pH7.4).

Storage Instruction: Store at +4℃ after thawing. Aliquot store at -20℃ or -80℃. Avoid repeated freeze / thaw cycles.

Purity: Protein A affinity purified.

Hangzhou Huaan Biotechnology Co., Ltd.

Orders:0086-571-88062880

Technical:0086-571-89986345

Service mail:support@huabio.cn

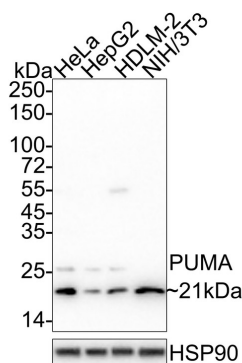
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Applications:WB=Western blot IHC-P=Immunohistochemistry (paraffin) IF-Cell=Immunofluorescence (Cell) IF-Tissue=Immunofluorescence (Tissue) FC=Flow cytometry IP=Immunoprecipitation

Images

Fig1: Western blot analysis of PUMA on different lysates with Rabbit anti-PUMA antibody (HA750038) at 1/2,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: HepG2 cell lysate
Lane 3: HDLM-2 cell lysate
Lane 4: NIH/3T3 cell lysate



Lysates/proteins at 20 µg/Lane.

Predicted band size: 21 kDa
Observed band size: 21 kDa

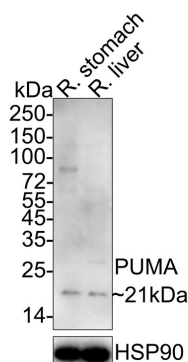
Exposure time: 24 seconds; ECL: K1801;

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 (HA750038) at 1/2,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

Fig2: Western blot analysis of PUMA on different lysates with Rabbit anti-PUMA antibody (HA750038) at 1/1,000 dilution.

Lane 1: Rat stomach tissue lysate
Lane 2: Rat liver tissue lysate



Lysates/proteins at 40 µg/Lane.

Predicted band size: 21 kDa
Observed band size: 21 kDa

Exposure time: 3 minutes; ECL: K1802;

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 (HA750038) at 1/1,000 dilution was used in 5% NFDM/TBST at 4°C overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

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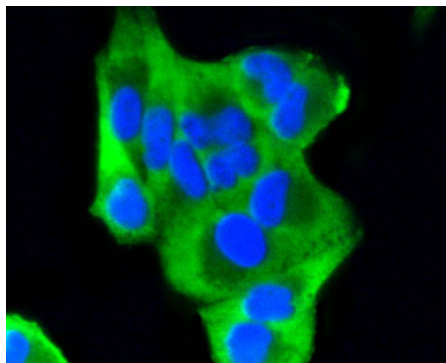


Fig3: ICC staining of PUMA in HeLa cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750038, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

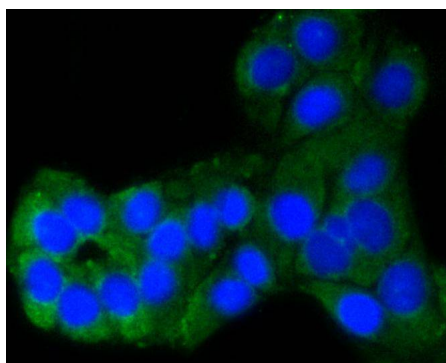


Fig4: ICC staining of PUMA in SKOV-3 cells (green). Formalin fixed cells were permeabilized with 0.1% Triton X-100 in TBS for 10 minutes at room temperature and blocked with 1% Blocker BSA for 15 minutes at room temperature. Cells were probed with the primary antibody (HA750038, 1/50) for 1 hour at room temperature, washed with PBS. Alexa Fluor®488 Goat anti-Rabbit IgG was used as the secondary antibody at 1/1,000 dilution. The nuclear counter stain is DAPI (blue).

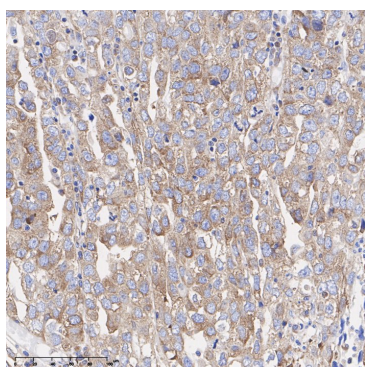


Fig5: Immunohistochemical analysis of paraffin-embedded human stomach cancer tissue with Rabbit anti-PUMA antibody (HA750038) at 1/5,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750038) at 1/5,000 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.

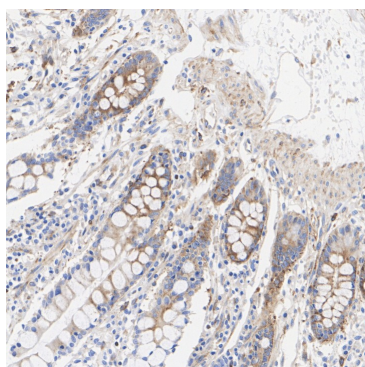


Fig6: Immunohistochemical analysis of paraffin-embedded human small intestine tissue with Rabbit anti-PUMA antibody (HA750038) at 1/10,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750038) at 1/10,000 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.

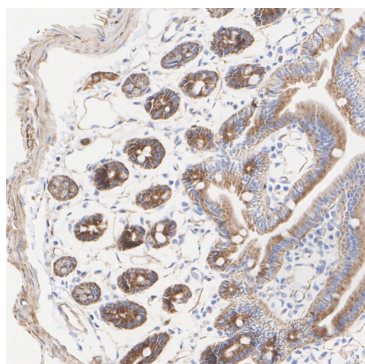


Fig7: Immunohistochemical analysis of paraffin-embedded mouse small intestine tissue with Rabbit anti-PUMA antibody (HA750038) at 1/30,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750038) at 1/30,000 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.

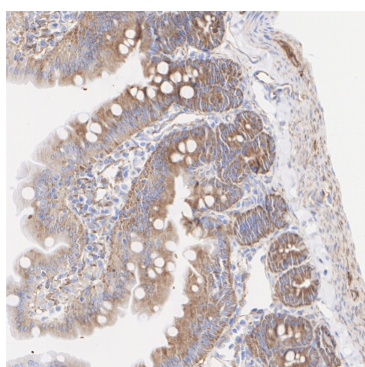


Fig8: Immunohistochemical analysis of paraffin-embedded rat small intestine tissue with Rabbit anti-PUMA antibody (HA750038) at 1/30,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750038) at 1/30,000 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.

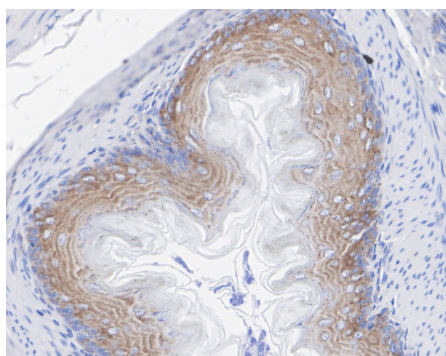


Fig9: Immunohistochemical analysis of paraffin-embedded rat stomach tissue with Rabbit anti-PUMA antibody (HA750038) at 1/2,000 dilution.

The section was pre-treated using heat mediated antigen retrieval with Tris-EDTA buffer (pH 9.0) for 20 minutes. The tissues were blocked in 1% BSA for 20 minutes at room temperature, washed with ddH₂O and PBS, and then probed with the primary antibody (HA750038) 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.

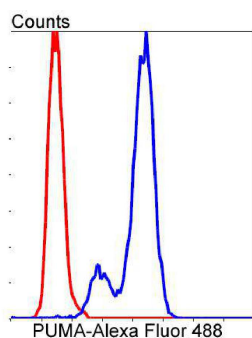


Fig10: Flow cytometric analysis of PUMA was done on HeLa cells. The cells were fixed, permeabilized and stained with the primary antibody (HA750038, 1/50) (blue). After incubation of the primary antibody at room temperature for an hour, the cells were stained with a Alexa Fluor 488-conjugated Goat anti-Rabbit IgG Secondary antibody at 1/1000 dilution for 30 minutes. Unlabelled sample was used as a control (cells without incubation with primary antibody; red).

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

Background References

1. Pace J et al. PuMA: A papillomavirus genome annotation tool. *Virus Evol.* 2020 Aug
2. Kennedy li DE et al. Contribution of Puma to Inflammatory Resolution During Early Pneumococcal Pneumonia. *Front Cell Infect Microbiol.* 2022 May

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