

## Anti-PUMA BBC3 Antibody

Catalog Number: A04899

### About BBC3

Apoptosis is related to many diseases and development. The p53 tumor-suppressor protein induces apoptosis through transcriptional activation of several genes. A novel p53 inducible pro-apoptotic gene was identified recently and designated PUMA (for p53 upregulated modulator of apoptosis) and bbc3 (for Bcl-2 binding component 3) in human and mouse. PUMA/bbc3 is one of the pro-apoptotic Bcl-2 family members including Bax and Noxa, which are also transcriptional targets of p53 (1). The PUMA gene encodes two BH3 domain-containing proteins termed PUMA-alpha and PUMA-beta (2). PUMA proteins bind Bcl-2, localize to the mitochondria, and induce cytochrome c release and apoptosis in response to p53. PUMA may be a direct mediator of p53-induced apoptosis.

### Overview

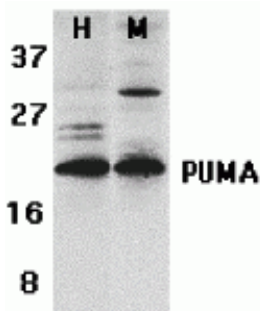
Product Name	Anti-PUMA BBC3 Antibody
Reactive Species	Human, Mouse
Description	Boster Bio Anti-PUMA BBC3 Antibody (Catalog # A04899). Tested in ELISA, WB, ICC, IF applications. This antibody reacts with Human, Mouse.
Application	ELISA, IF, ICC, WB
Clonality	Polyclonal
Formulation	PUMA Antibody is supplied in PBS containing 0.02% sodium azide.
Storage Instructions	PUMA antibody can be stored at 4°C for three months and -20°C, stable for up to one year. Avoid repeated freeze-thaw cycles. Antibodies should not be exposed to prolonged high temperatures.
Host	Rabbit
Uniprot ID	Q96PG8

### Technical Details

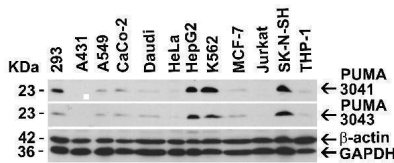
Immunogen	Anti-PUMA antibody was raised against a peptide corresponding to 14 amino acids near the carboxyl terminus human PUMA isoform 1. The immunogen is located within the last 50 amino acids of PUMA.
Predicted Reactive Species	Rat
Cross Reactivity	A lower band at approximately 16 kDa was detected in Daudi and K562 cells, which may represent the PUMA isoform 2.
Isotype	IgG
Form	Liquid

Concentration	1 mg/mL
Purification	PUMA Antibody is affinity chromatography purified via peptide column.
Suggested Dilutions	WB: 1-4 ug/mL; IF: 2 ug/mL; ICC: 1 ug/mL. Antibody validated: Western Blot in human and mouse samples; Immunocytochemistry and Immunofluorescence in human samples. All other applications and species not yet tested. Optimal dilutions for each application should be determined by the researcher.

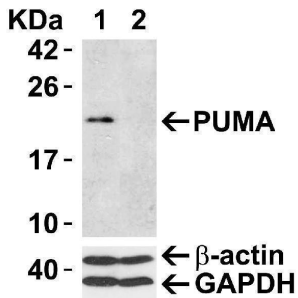
## Anti-PUMA BBC3 Antibody (A04899) Images



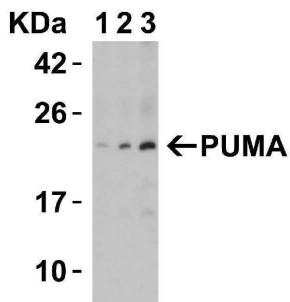
Western Blot Validation of PUMA in K562 and 3T3/NIH Cells  
Loading: 15 ug of lysates per lane. Antibodies: A04899 (2 ug/mL), 1 h incubation at RT in 5% NFD/MTBST. Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



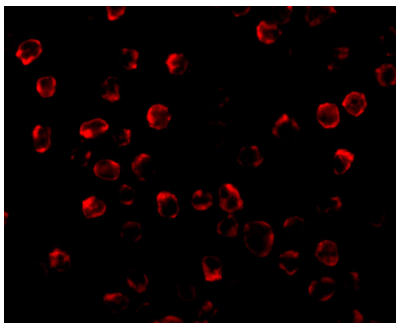
Independent Antibody Validation (IAV) via Protein Expression Profile in Cell Lines Loading: 20 ug of lysates per lane. Antibodies: A04899 (3 ug/mL), 3043 (2 ug/mL), beta-actin (1 ug/mL) and GAPDH (0.02 ug/mL), 1 h incubation at RT in 5% NFD/MTBST. Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



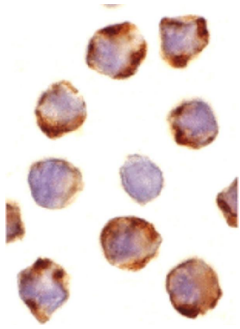
Validation with PUMA siRNA Knockdown in 293 Cells 293 cells were transfected with control siRNAs (lane 1) or PUMA siRNAs (lane 2) Loading: 15 ug of 293 whole cell lysates per lane. Antibodies: A04899 (2 ug/mL), beta-actin (1 ug/mL) and GAPDH (0.02 ug/mL), 1 h incubation at RT in 5% NFD/MTBST. Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



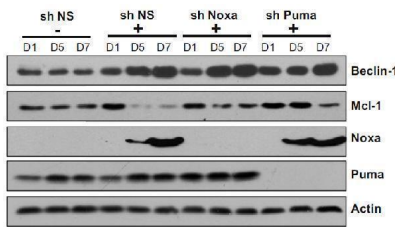
Sensitivity Test for PUMA in 2983 Cells Loading: Lysates/proteins at 15 ug per lane. Antibodies: A04899 (lane 1-3: 1, 2 and 4 ug/mL). 1 h incubation at RT in 5% NFD/MTBST. Secondary: Goat anti-rabbit IgG HRP conjugate at 1:10000 dilution.



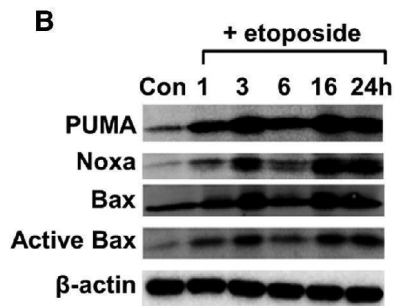
Immunofluorescence Validation of PUMA in K562 Cells  
Immunofluorescent analysis of 4% paraformaldehyde-fixed K562 cells labeling PUMA with A04899 at 2 ug/mL, followed by goat anti-rabbit IgG secondary antibody at 1/500 dilution (red). Image showing cytosol staining on K562 cells.



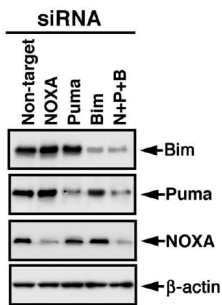
**Immunocytochemistry Validation of PUMA in K562 Cells**  
Immunocytochemical analysis of K562 cells using anti-PUMA antibody (A04899) at 1 ug/ml. Cells was fixed with formaldehyde and blocked with 10% serum for 1 h at RT; antigen retrieval was by heat mediation with a citrate buffer (pH6). Samples were incubated with primary antibody overnight at 4°C. A goat anti-rabbit IgG H&L (HRP) at 1/250 was used as secondary. Counter stained with Hematoxylin.



**KD Validation of PUMA in HOSE-RasV12 Cells** (Elgendy et al., 2011) HOSE-RasV12 cells were transfected with control shRNA plasmid or shRNA plasmids (KD) targeted against Noxa or Puma, as indicated. PUMA expression was not observed in PUMA KD cells detected by anti-PUMA antibodies (A04899).



**Induction Validation of PUMA in Primary Cortical Neurons** (Sabirzhanov et al., 2014) PUMA protein levels were increased in etoposide-treated primary cortical neurons detected by anti-PUMA antibodies (A04899).



**KD Validation of PUMA PUMA in Tet Cells** (Han et al., 2010) Immunoblot analyses of Tet-induced p53 cells treated with NOXA, Puma, Bim or non-targeting siRNAs that were utilized in this experiment. PUMA protein levels were markedly reduced in PUMA KD cells detected by anti-PUMA antibodies (A04899).

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Anti-PUMA BBC3 Antibody

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