

## Anti-Smac/Diablo Antibody Picoband®

Catalog Number: PB9860

### About DIABLO

Diablo homolog (DIABLO), also referred to as second mitochondria-derived activator of caspases or SMAC, is a mitochondrial protein that in humans is encoded by the DIABLO (direct IAP binding protein with low pI) gene on chromosome 12. The encoded mitochondrial protein enters the cytosol when cells undergo apoptosis, and allows activation of caspases by binding to inhibitor of apoptosis proteins. Overexpression of the encoded protein sensitizes tumor cells to apoptosis. A mutation in this gene is associated with young-adult onset of nonsyndromic deafness-64. Alternative splicing results in multiple transcript variants encoding different isoforms.

### Overview

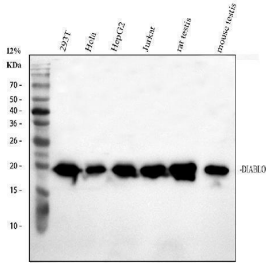
Product Name	Anti-Smac/Diablo Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-Smac/Diablo Antibody Picoband® catalog # PB9860. Tested in ELISA, Flow Cytometry, IP, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a premium antibody that guarantees superior quality, high affinity, and strong signals with minimal background in Western blot applications. Only our best-performing antibodies are designated as Picoband, ensuring unmatched performance.
Application	ELISA, Flow Cytometry, IP, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains 4 mg Trehalose, 0.9 mg NaCl and 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> .
Storage Instructions	Store at -20°C for one year from date of receipt. After reconstitution, at 4°C for one month. It can also be aliquotted and stored frozen at -20°C for six months. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	Q9NR28

### Technical Details

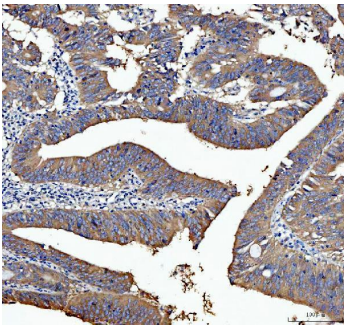
Immunogen	E. coli-derived human Smac/Diablo recombinant protein (Position: A56-D239). Human Smac/Diablo shares 88.6% amino acid (aa) sequence identity with mouse Smac/Diablo.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit IgG Super Vision Assay Kit (SV0002-1) for IHC(P) and ICC.
Cross Reactivity	No cross-reactivity with other proteins.
Isotype	Rabbit IgG
Form	Lyophilized

Concentration	Adding 0.2 ml of distilled water will yield a concentration of 500 ug/ml.
Purification	Immunogen affinity purified.
Suggested Dilutions	Western blot, 0.1-0.5ug/ml, Human, Mouse, Rat Immunohistochemistry (Paraffin-embedded Section), 2-5ug/ml, Human Immunocytochemistry/Immunofluorescence, 5ug/ml, Human Immunoprecipitation, 0.5-2 ug/ml, Human Flow Cytometry(Fixed), 1-3 ug/1x10 <sup>6</sup> cells, Human ELISA, 0.1-0.5ug/ml, -

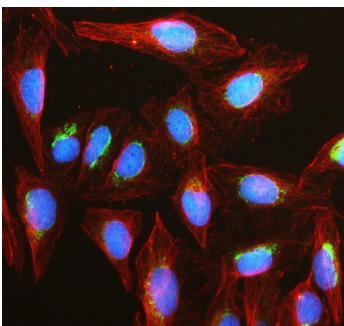
## Anti-Smac/Diablo Antibody Picoband® (PB9860) Images



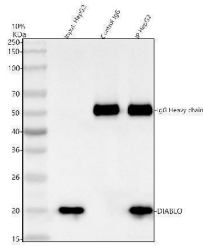
Western blot analysis of Smac/Diablo using anti-Smac/Diablo antibody (PB9860). Electrophoresis was performed on a 12 SDS-PAGE gel at 80V (Stacking gel) / 120V (Resolving gel) for 2 hours. The sample well of each lane was loaded with 30 ug of sample under reducing conditions. Lane 1: human 293T whole cell lysates, Lane 2: human Hela whole cell lysates, Lane 3: human HepG2 whole cell lysates, Lane 4: human Jurkat whole cell lysates, Lane 5: rat testis tissue lysates, Lane 6: mouse testis tissue lysates. After electrophoresis, proteins were transferred to a nitrocellulose membrane at 150 mA for 50-90 minutes. Blocked the membrane with 5% non-fat milk/TBS for 1.5 hour at RT. The membrane was incubated with rabbit anti-Smac/Diablo antigen affinity purified polyclonal antibody (Catalog # PB9860) at 0.5 ug/mL overnight at 4°C, then washed with TBS-0.1%Tween 3 times with 5 minutes each and probed with a goat anti-rabbit IgG-HRP secondary antibody at a dilution of 1:5000 for 1.5 hour at RT. The signal is developed using an ECL Plus Western Blotting Substrate (Catalog # AR1196-200) with Tanon 5200 system. A specific band was detected for Smac/Diablo at approximately 20 kDa. The expected band size for Smac/Diablo is at 27 kDa.



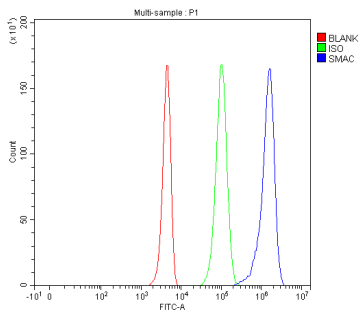
IHC analysis of Smac/Diablo using anti-Smac/Diablo antibody (PB9860). Smac/Diablo was detected in a paraffin-embedded section of human rectal cancer tissue. Heat mediated antigen retrieval was performed in EDTA buffer (pH 8.0, epitope retrieval solution). The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 2 ug/ml rabbit anti-Smac/Diablo Antibody (PB9860) overnight at 4°C. Peroxidase Conjugated Goat Anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using HRP Conjugated Rabbit IgG Super Vision Assay Kit (Catalog # SV0002) with DAB as the chromogen.



IF analysis of Smac/Diablo using anti-Smac/Diablo antibody (PB9860) and anti-Tubulin Alpha antibody (M03989-3). Smac/Diablo was detected in immunocytochemical section of U2OS cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 5 ug/mL rabbit anti-Smac/Diablo Antibody (PB9860) and mouse anti-Tubulin Alpha antibody (M03989-3) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) and Cy3 Conjugated Goat Anti-Mouse IgG (BA1031) were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. The section was counterstained with DAPI. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



Immunoprecipitating (IP) Smac/Diablo in HepG2 whole cell lysate. Western blot analysis of Smac/Diablo using anti-Smac/Diablo antibody (PB9860); Lane 1: HepG2 whole cell lysates (30ug); Lane 2: Rabbit control IgG instead of anti-Smac/Diablo antibody in HepG2 whole cell lysate; Lane 3: anti-Smac/Diablo antibody (2ug) + HepG2 whole cell lysate (500ug). After electrophoresis, proteins were transferred to a membrane. Then the membrane was incubated with rabbit anti-Smac/Diablo antigen affinity purified polyclonal antibody (PB9860) at a dilution of 0.5 ug/mL and probed with a goat anti-rabbit IgG-HRP secondary antibody (Catalog # BA1054). The signal is developed using ECL Plus Western Blotting Substrate (Catalog # AR1196-200). A specific band was detected for Smac/Diablo at approximately 20 kDa. The expected band size for Smac/Diablo is at 27 kDa.



Flow Cytometry analysis of 293T cells using anti-Smac/Diablo antibody (PB9860). Overlay histogram showing 293T cells stained with PB9860 (Blue line). To facilitate intracellular staining, cells were fixed with 4% paraformaldehyde and permeabilized with permeabilization buffer. The cells were blocked with 10% normal goat serum. And then incubated with rabbit anti-Smac/Diablo Antibody (PB9860, 1 ug/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit IgG (BA1127, 5-10 ug/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1 ug/1x10<sup>6</sup>) used under the same conditions. Unlabelled sample without incubation with primary antibody and secondary antibody (Red line) was used as a blank control.

## 1 Publications Citing This Product

1. PubMed ID: 28081713, You F, Li Q, Jin G, Zheng Y, Chen J, Yang H. BMC Neurosci. 2017 Jan 12;18(1):12. doi: 10.1186/s12868-016-0329-9. Genistein protects against Abeta25–35 induced apoptosis of PC12 cells through JNK signaling and modulation of Bcl-2 family messengers

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