

## Anti-MAD1/MAD1L1 Antibody Picoband®

Catalog Number: PB9262

### About MAD1L1

Mitotic spindle assembly checkpoint protein MAD1 is a protein that in humans is encoded by the MAD1L1 gene. It is mapped to 7p22.3. MAD1L1 is a component of the mitotic spindle-assembly checkpoint that prevents the onset of anaphase until all chromosome are properly aligned at the metaphase plate. MAD1L1 can function as a homodimer. It localizes to the centrosome during metaphase and to the spindle midzone and the midbody during anaphase and telophase. MAD1L1 may also play a role in cell cycle control and tumor suppression.

### Overview

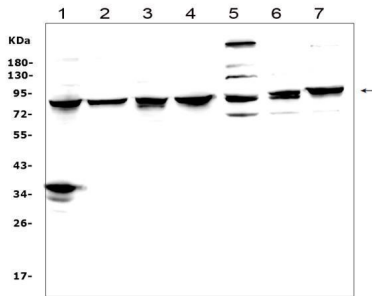
Product Name	Anti-MAD1/MAD1L1 Antibody Picoband®
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-MAD1/MAD1L1 Antibody Picoband® catalog # PB9262. Tested in Flow Cytometry, IF, IHC, ICC, WB applications. This antibody reacts with Human, Mouse, Rat. The brand Picoband indicates this is a 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	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Polyclonal
Formulation	Each vial contains antibody formulated with stabilizing components, 0.9 mg NaCl, 0.2 mg Na <sub>2</sub> HPO <sub>4</sub> , and 0.05 mg NaN <sub>3</sub> . *This antibody is supplied in a stabilized formulation. Compatibility with conjugation reactions depends on the chemistry of the conjugation method used. For conjugation methods that are not compatible with the stabilizing components present in this formulation, a carrier-free antibody format is required.
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	Q9Y6D9

### Technical Details

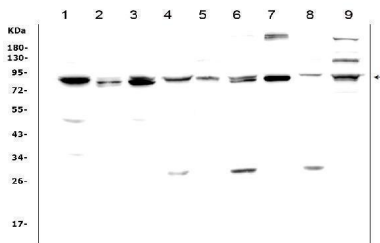
Immunogen	E.coli-derived human MAD1 recombinant protein (Position: L362-A632). Human MAD1 shares 81% amino acid (aa) sequence identity with mouse MAD1.
Recommended Detection Systems	Boster recommends Enhanced Chemiluminescent Kit with anti-Rabbit IgG (EK1002) for Western blot, and HRP Conjugated anti-Rabbit 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), 0.5-1ug/ml, Human, Mouse, Rat Immunocytochemistry, 0.5-1ug/ml, Human Immunocytochemistry/Immunofluorescence, 2ug/ml, Human Flow Cytometry (Fixed), 1-3ug/1x10 <sup>6</sup> cells, Human

## Anti-MAD1/MAD1L1 Antibody Picoband® (PB9262) Images

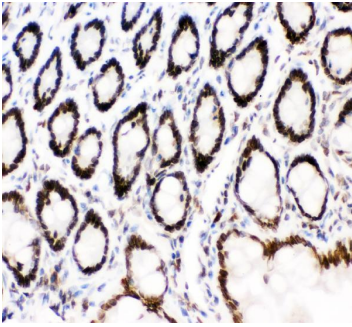


Western blot analysis of MAD1 using anti-MAD1 antibody (PB9262). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions. Lane 1: human HeLa whole cell lysates, Lane 2: human Raji whole cell lysates, Lane 3: human Jurkat whole cell lysates, Lane 4: human HepG2 whole cell lysates. Lane 5: human HL-60 whole cell lysates. Lane 6: human THP-1 whole cell lysates. Lane 7: human Caco-2 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-MAD1 antigen affinity purified polyclonal antibody (Catalog # PB9262) 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 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for MAD1 at approximately 83KD. The expected band size for MAD1 is at 83KD.

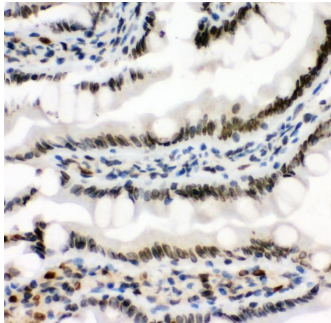


Western blot analysis of MAD1 using anti-MAD1 antibody (PB9262). Electrophoresis was performed on a 5-20% SDS-PAGE gel at 70V (Stacking gel) / 90V (Resolving gel) for 2-3 hours. The sample well of each lane was loaded with 50ug of sample under reducing conditions. Lane 1: rat brain tissue lysates, Lane 2: rat kidney tissue lysates, Lane 3: rat lung tissue lysates, Lane 4: rat liver tissue lysates, Lane 5: mouse brain tissue lysates, Lane 6: mouse kidney tissue lysates, Lane 7: mouse lung tissue lysates, Lane 8: mouse liver tissue lysates, Lane 9: mouse NIH3T3 whole cell lysates. After Electrophoresis, proteins were transferred to a Nitrocellulose membrane at 150mA 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-MAD1 antigen affinity purified polyclonal antibody (Catalog # PB9262) 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 1:10000 for 1.5 hour at RT. The signal is developed using an Enhanced Chemiluminescent detection (ECL) kit (Catalog # EK1002) with Tanon 5200 system. A specific band was detected for MAD1 at approximately 83KD. The expected band size for MAD1 is at 83KD.

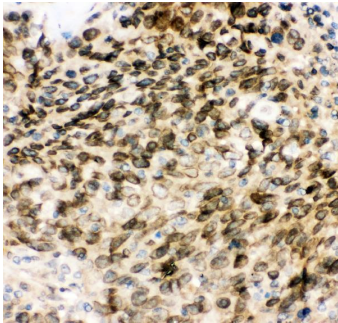
IHC analysis of MAD1 using anti-MAD1 antibody (PB9262). MAD1 was detected in paraffin-embedded section of mouse intestine tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat



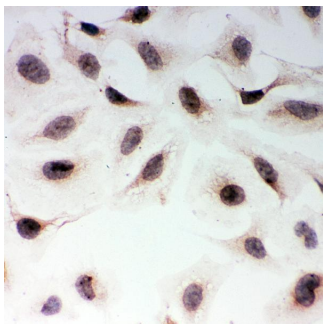
serum. The tissue section was then incubated with 1ug/ml rabbit anti-MAD1 Antibody (PB9262) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.



IHC analysis of MAD1 using anti-MAD1 antibody (PB9262). MAD1 was detected in paraffin-embedded section of rat intestine tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-MAD1 Antibody (PB9262) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

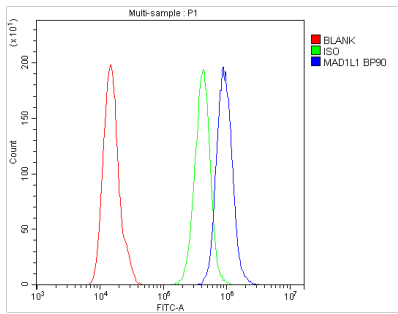


IHC analysis of MAD1 using anti-MAD1 antibody (PB9262). MAD1 was detected in paraffin-embedded section of human intestinal cancer tissues. Heat mediated antigen retrieval was performed in citrate buffer (pH6, epitope retrieval solution) for 20 mins. The tissue section was blocked with 10% goat serum. The tissue section was then incubated with 1ug/ml rabbit anti-MAD1 Antibody (PB9262) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The tissue section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

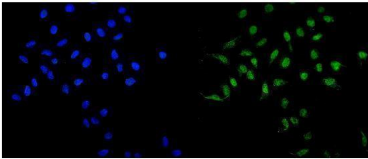


IHC analysis of MAD1 using anti-MAD1 antibody (PB9262). MAD1 was detected in immunocytochemical section of Hela cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated with 1ug/ml rabbit anti-MAD1 Antibody (PB9262) overnight at 4°C. Biotinylated goat anti-rabbit IgG was used as secondary antibody and incubated for 30 minutes at 37°C. The section was developed using Streptavidin-Biotin-Complex (SABC)(Catalog # SA1022) with DAB as the chromogen.

Flow Cytometry analysis of A431 cells using anti-MAD1 antibody (PB9262). Overlay histogram showing A431 cells stained with PB9262 (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-MAD1 Antibody (PB9262, 1ug/1x10<sup>6</sup> cells) for 30 min at 20°C. DyLight®488 conjugated goat anti-rabbit



IgG (BA1127, 5-10ug/1x10<sup>6</sup> cells) was used as secondary antibody for 30 minutes at 20°C. Isotype control antibody (Green line) was rabbit IgG (1ug/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.



IF analysis of MAD1 using anti-MAD1 antibody (PB9262). MAD1 was detected in immunocytochemical section of U2OS cells. 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 2ug/mL rabbit anti-MAD1 Antibody (PB9262) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) was used as secondary antibody at 1:100 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.

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### Anti-MAD1/MAD1L1 Antibody

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