

Anti-HMGB1/Hmg 1 Rabbit Monoclonal Antibody

Catalog Number: M00066-1

About HMGB1

Phosphoinositide-3-kinase (PI3K) that phosphorylates PtdIns (Phosphatidylinositol), PtdIns4P (Phosphatidylinositol 4-phosphate) and PtdIns (4,5) P2 (Phosphatidylinositol 4,5-bisphosphate) to generate phosphatidylinositol 3,4,5-trisphosphate (PIP3) . PIP3 plays a key role by recruiting PH domain-containing proteins to the membrane, including AKT1 and PDPK1, activating signaling cascades involved in cell growth, survival, proliferation, motility and morphology. Participates in cellular signaling in response to various growth factors.

Overview

Product Name	Anti-HMGB1/Hmg 1 Rabbit Monoclonal Antibody
Reactive Species	Human, Mouse, Rat
Description	Boster Bio Anti-HMGB1/Hmg 1 Rabbit Monoclonal Antibody catalog # M00066-1. Tested in WB, IHC, ICC/IF, Flow Cytometry applications. This antibody reacts with Human, Mouse, Rat.
Application	Flow Cytometry, IF, IHC, ICC, WB
Clonality	Monoclonal AAC-8
Formulation	Rabbit IgG in phosphate buffered saline, pH 7.4, 150mM NaCl, 0.02% sodium azide and 50% glycerol, 0.4-0.5mg/ml BSA.
Storage Instructions	Store at -20°C for one year. For short term storage and frequent use, store at 4°C for up to one month. Avoid repeated freeze-thaw cycles.
Host	Rabbit
Uniprot ID	P09429

Technical Details

Immunogen	A synthesized peptide derived from human HMGB1
Isotype	Rabbit IgG
Form	Liquid
Concentration	Actual concentration vary by lot. Use suggested dilution ratio to decide dilution procedure.
Purification	Affinity-chromatography
Suggested Dilutions	Dilute the sample so that the expected range of concentrations fall within the detection range of this kit. If the expected range of concentration is unknown, a pilot test should be conducted to decide the optimal dilution ratio for your samples. Some PubMed article(s) citing the expression level of this target are as follows:

Boster Bio's internal QC testing used:

WB 1:1000-1:2000

IHC 1:50-1:100

ICC/IF 1:50-1:100

FC 1:30

Anti-HMGB1/Hmg 1 Rabbit Monoclonal Antibody (M00066-1) Images

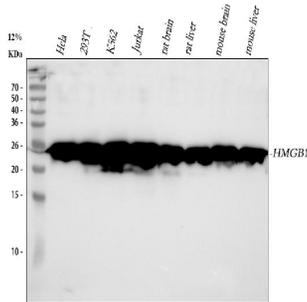


Figure 1. Western blot analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

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 30 ug of sample under reducing conditions.

Lane 1: human Hela whole cell lysates,
Lane 2: human 293T whole cell lysates,
Lane 3: human K562 whole cell lysates,
Lane 4: human Jurkat whole cell lysates,
Lane 5: rat brain tissue lysates,
Lane 6: rat liver tissue lysates,
Lane 7: mouse brain tissue lysates,
Lane 8: mouse liver 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-HMGB1 antigen affinity purified monoclonal antibody (Catalog # M00066-1) at 1:1000 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 1:1000 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 HMGB1 at approximately 25 kDa. The expected band size for HMGB1 is at 25 kDa.

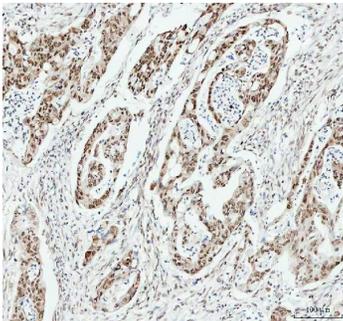


Figure 2. IHC analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

HMGB1 was detected in a paraffin-embedded section of human colorectal adenocarcinoma 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 1:50 rabbit anti-HMGB1 Antibody (M00066-1) 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.

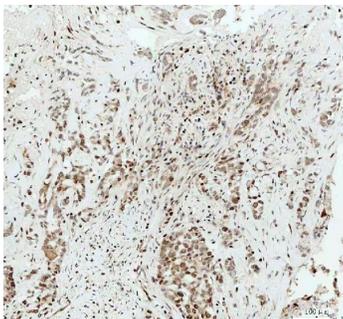


Figure 3. IHC analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

HMGB1 was detected in a paraffin-embedded section of human lung adenocarcinoma 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 1:50 rabbit anti-HMGB1 Antibody (M00066-1) 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.



Figure 4. IHC analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

HMGB1 was detected in a paraffin-embedded section of human spleen 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 1:50 rabbit anti-HMGB1 Antibody (M00066-1) 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.

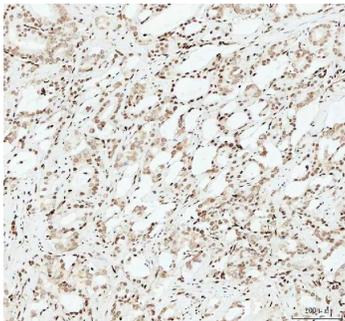


Figure 5. IHC analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

HMGB1 was detected in a paraffin-embedded section of human prostatic acinar adenocarcinoma 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 1:50 rabbit anti-HMGB1 Antibody (M00066-1) 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.

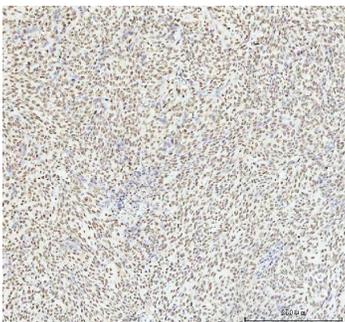


Figure 6. IHC analysis of HMGB1 using anti-HMGB1 antibody (M00066-1).

HMGB1 was detected in a paraffin-embedded section of human cervical squamous carcinoma 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 1:50 rabbit anti-HMGB1 Antibody (M00066-1) 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.

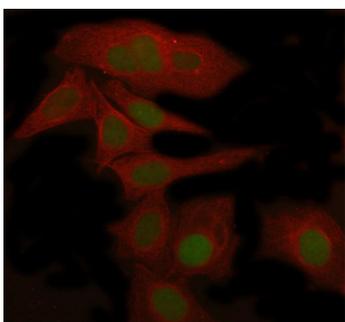
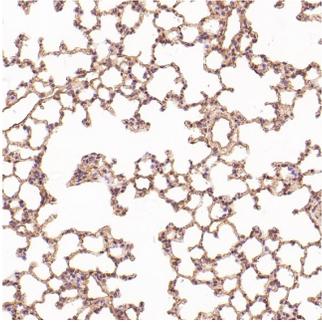
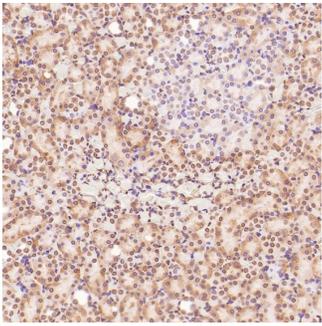


Figure 7. IF analysis of HMGB1 using anti-HMGB1 antibody (M00066-1) and anti-Beta Tubulin antibody (M01857-3). HMGB1 was detected in immunocytochemical section of HeLa cell. Enzyme antigen retrieval was performed using IHC enzyme antigen retrieval reagent (AR0022) for 15 mins. The cells were blocked with 10% goat serum. And then incubated at 1:50 with rabbit anti-HMGB1 Antibody (M00066-1) and mouse anti-Beta Tubulin antibody (M01857-3) overnight at 4°C. DyLight®488 Conjugated Goat Anti-Rabbit IgG (BA1127) and Cy3 Conjugated Goat Anti-

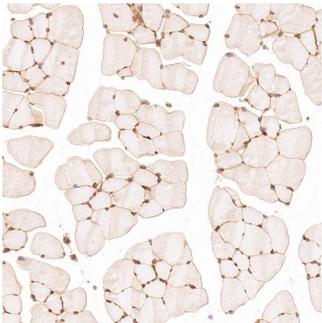
Mouse IgG (BA1031) were used as secondary antibody at 1:500 dilution and incubated for 30 minutes at 37°C. Visualize using a fluorescence microscope and filter sets appropriate for the label used.



Immunohistochemical analysis of paraffin-embedded Rat liver, using the Antibody at 1:300 dilution.



Immunohistochemical analysis of paraffin-embedded Rat kidney, using the Antibody at 1:300 dilution.



Immunohistochemical analysis of paraffin-embedded Mouse skeletal muscle - gastrocnemius , using the Antibody at 1:300 dilution.

5 Publications Citing This Product

1. PubMed ID: 31933513, Wen Y,Sun HY,Tan Z,Liu RH,Huang SQ,Chen GY,Qi H,Tang LJ.Abdominal paracentesis drainage ameliorates myocardial injury in severe experimental pancreatitis rats through suppressing oxidative stress.World J Gastroenterol.2020 Jan 7;26(1):35-54.doi:10.3748/wj
2. PubMed ID: 25685165, Association of Upregulated HMGB1 and c-IAP2 Proteins With Hepatocellular Carcinoma Development and Progression
3. PubMed ID: 26648090, Galantamine protects against lipopolysaccharide-induced acute lung injury in rats

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