Anti-BAX Antibody

ER0907

Product Type:	Rabbit polyclonal IgG, primary antibodies
Species reactivity:	Human, Mouse, Rat
Applications:	WB, IF-Cell, IHC-P
Molecular Wt:	Predicted band size: 21 kDa

Description: BAX is a member of the Bcl-2 gene family. Apoptosis regulator BAX promotes apoptosis by binding to and antagonizing the Bcl-2 protein. In healthy mammalian cells, the majority of BAX is found in the cytosol, but upon initiation of apoptotic signaling, Bax undergoes a conformational shift. Upon induction of apoptosis, BAX becomes organelle membrane-associated, and in particular, mitochondrial membrane associated. The expression of BAX is upregulated by the tumor suppressor protein p53, and BAX has been shown to be involved in p53-mediated apoptosis. The p53 protein is a transcription factor that, when activated as part of the cell's response to stress, regulates many downstream target genes, including BAX.

Immunogen: Synthetic peptide within Human BAX aa 1-50 / 192.

Positive control: HeLa cell lysate, MCF7 cell lysate, HEK-293 cell lysate, Daudi cell lysate, Raji cell lysate, Jurkat cell lysate, HepG2, F9, SHSY5Y, human colon carcinoma tissue, mouse kidney tissue.

Subcellular location: Mitochondrion membrane, cytoplasm.

Database links: SwissProt: Q07812 Human | Q07813 Mouse | Q63690 Rat

Recommended Dilutions:

WB	1:5,000-1:20,000
IF-Cell	1:200
IHC-P	1:1,000

Storage Buffer: 1*PBS (pH7.4), 0.2% BSA, 40% Glycerol. Preservative: 0.05% Sodium Azide.

Storage Instruction: Shipped at 4℃. Store at +4℃ short term (1-2 weeks). It is recommended to aliquot into single-use upon delivery. Store at -20℃ long term.

Purity: Immunogen affinity purified.

Images

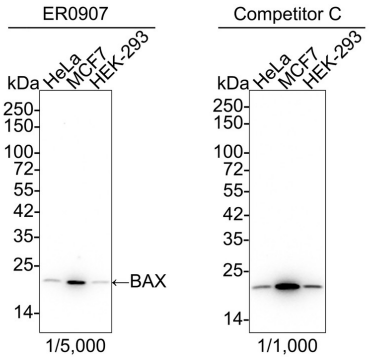

Fig1: Western blot analysis of BAX on different lysates with Rabbit anti-BAX antibody (ER0907) at 1/5,000 dilution and competitor's antibody at 1/1,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: MCF7 cell lysate
Lane 3: HEK-293 cell lysate

Lysates/proteins at 15 µg/Lane.

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

Exposure time: 1 minute 34 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 (ER0907) at 1/5,000 dilution and competitor's antibody at 1/1,000 dilution were used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1/50,000 dilution was used for 1 hour at room temperature.

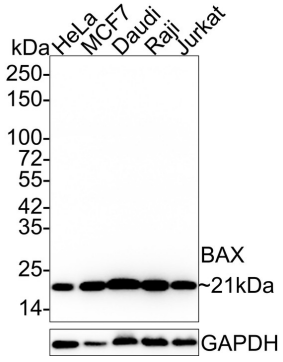

Fig2: Western blot analysis of BAX on different lysates with Rabbit anti-BAX antibody (ER0907) at 1/5,000 dilution.

Lane 1: HeLa cell lysate
Lane 2: MCF7 cell lysate
Lane 3: Daudi cell lysate
Lane 4: Raji cell lysate
Lane 5: Jurkat cell lysate

Lysates/proteins at 20 µg/Lane.

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

Exposure time: 29 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 (ER0907) at 1/5,000 dilution was used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-Rabbit IgG - HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

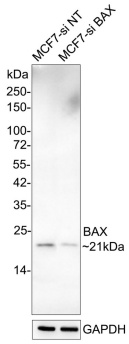

Fig3: Western blot analysis of BAX on different lysates with Rabbit anti-BAX antibody (ER0907) at 1/20,000 dilution.

Lane 1: MCF7-si NT cell lysate
Lane 2: MCF7-si BAX cell lysate

Lysates/proteins at 10 µg/Lane.

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

Exposure time: 3 minutes; ECL: K1802;
4-20% SDS-PAGE gel.

ER0907 was shown to specifically react with BAX in MCF7-si NT cells. Weakened band was observed when MCF7-si BAX sample was tested. MCF7-si NT and MCF7-si BAX samples were subjected to SDS-PAGE. Proteins were transferred to a PVDF membrane and blocked with 5% NFDM in TBST for 1 hour at room temperature. The primary antibody (ER0907, 1/20,000) and Loading control antibody (Rabbit anti-GAPDH, ET1601-4, 1/10,000) were used in 5% NFDM/TBST at 4℃ overnight. Goat Anti-rabbit IgG-HRP Secondary Antibody (HA1001) at 1:50,000 dilution was used for 1 hour at room temperature.

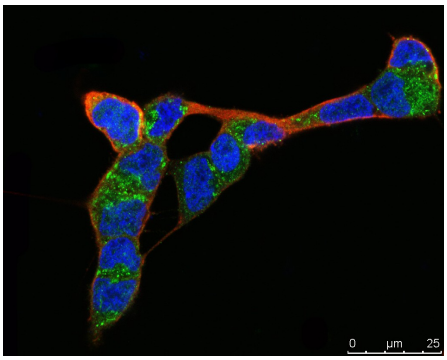

Fig4: Immunocytochemistry analysis of SHSY5Y cells labeling BAX with Rabbit anti-BAX antibody (ER0907) at 1/200 dilution.

Cells were fixed in 4% paraformaldehyde for 10 minutes at 37 ℃, permeabilized with 0.05% Triton X-100 in PBS for 20 minutes, and then blocked with 2% negative goat serum for 30 minutes at room temperature. Cells were then incubated with Rabbit anti-BAX antibody (ER0907) at 1/200 dilution in 2% negative goat serum overnight at 4 ℃. Goat Anti-Rabbit IgG H&L (iFluor™ 488, HA1121) was used as the secondary antibody at 1/1,000 dilution. Nuclear DNA was labelled in blue with DAPI.

Beta III tubulin (M1305-2, red) was stained at 1/200 dilution overnight at +4℃. Goat Anti-Mouse IgG H&L (iFluor™ 647, HA1127) were used as the secondary antibody at 1/1,000 dilution.

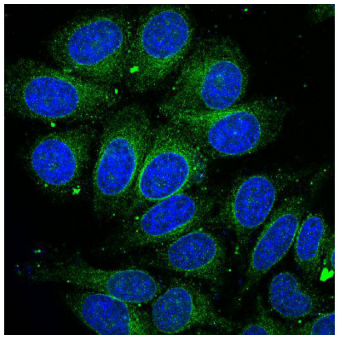

Fig5: ICC staining Bax in HepG2 cells (green). The nuclear counter stain is DAPI (blue).Cells were fixed in paraformaldehyde, permeabilised with 0.25% Triton X100/PBS.

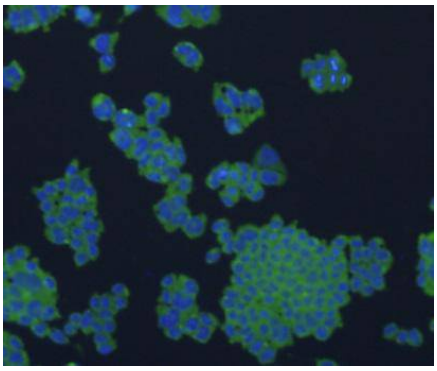

Fig6: ICC staining Bax in F9 cells (green). The nuclear counter stain is DAPI (blue). Cells were fixed in paraformaldehyde, permeabilised with 0.25% Triton X100/PBS.

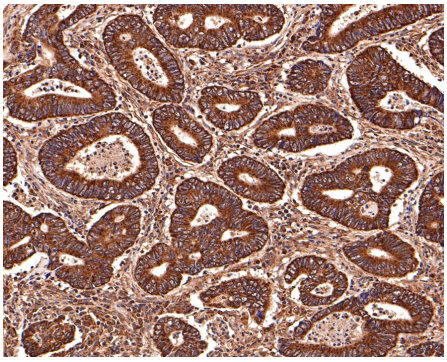

Fig7: Immunohistochemical analysis of paraffin-embedded human colon carcinoma tissue with Rabbit anti-BAX antibody (ER0907) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (ER0907) at 1/1,000 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.

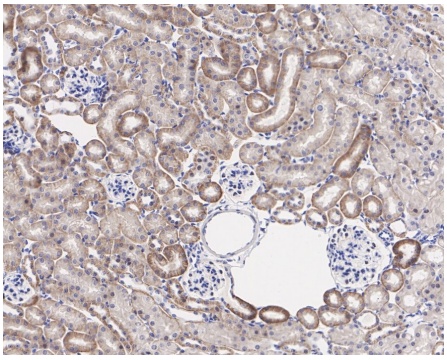

Fig8: Immunohistochemical analysis of paraffin-embedded mouse kidney tissue with Rabbit anti-BAX antibody (ER0907) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (ER0907) at 1/1,000 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.

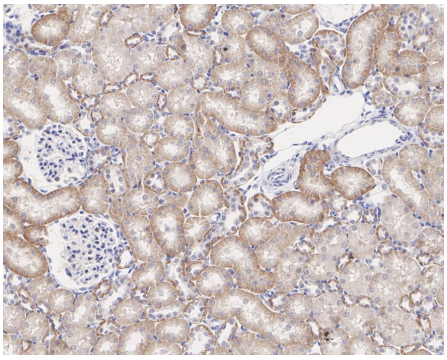

Fig9: Immunohistochemical analysis of paraffin-embedded rat kidney tissue with Rabbit anti-BAX antibody (ER0907) at 1/1,000 dilution.

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 ddH_2O and PBS, and then probed with the primary antibody (ER0907) at 1/1,000 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.

Background References

1. "Elucidation of some Bax conformational changes through crystallization of an antibody-peptide complex." Peyerl F.W., Dai S., Murphy G.A., Crawford F., White J., Marrack P., Kappler J.W. Cell Death Differ. 14:447-452(2006)
2. "BAX activation is initiated at a novel interaction site." Gavathiotis E., Suzuki M., Davis M.L., Pitter K., Bird G.H., Katz S.G., Tu H.C., Kim H., Cheng E.H., Tjandra N., Walensky L.D. Nature 455:1076-1081(2007)
3. "Mutation to Bax beyond the BH3 domain disrupts interactions with pro-survival proteins and promotes apoptosis." Czabotar P.E., Lee E.F., Thompson G.V., Wardak A.Z., Fairlie W.D., Colman P.M. J. Biol. Chem. 286:7123-7131(2010)