

# TREVIGEN® Product Data

*For Research Use Only. Not For Use In Diagnostic Procedures*

## Anti-Bax YTH-5B7 Monoclonal Antibody

**Catalog #:** 2280-MC-100

**Size:** 100 µg

**Description:** The Bcl-2 family of proteins plays a crucial role in the regulation of cell death in many eukaryotic systems. Bax has been shown to redistribute from the cytosol to the mitochondria during apoptosis, and over-expression of Bax can accelerate cell death. Coregulation of Bax dimer formation and intracellular localization are associated with Bax conformational changes. When used in conjunction with the anti-Bax YTH-6A7 (cat# 2281-MC-100), which recognizes only detergent-treated Bax, the researcher may selectively identify the activated form of Bax.

**Physical State:** This antibody is provided as purified immunoglobulin from mouse ascites in 1X PBS containing 0.01% sodium azide.

**Immunogen:** A synthetic peptide corresponding to amino acids 3 to 16 of the mouse Bax sequence.

**Ig Class:** IgG<sub>1</sub>

**Specificity:** Anti-Bax YTH-5B7 is specific for mouse and does not cross react with human Bax.

**Storage:** Store at 4°C.

**Applications:** Western analysis, and immunoprecipitation. For Western blots, an antibody dilution of 1:1000 is recommended.

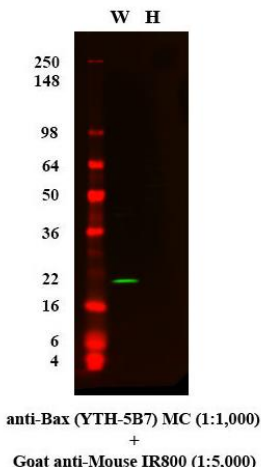


Fig. 1. Western blot analysis of mouse Wehi and human HT1080 cell lines. Cells were lysed in Tris-Glycine SDS sample buffer at a concentration of  $1 \times 10^7$  cells/ml, and 10 µl of lysates were loaded per well of a 4-20% Tris-Glycine gel. Proteins were transferred onto an Immobilon FL membrane, detected using Trevigen's anti-Bax (YTH-5B7) antibody (cat# 2280-MC-100) and visualized using an IR800-conjugated secondary antibody. The membrane was scanned using an Odyssey Infrared Imaging System (Licor). Trevigen's Bax antibody (clone YTH-5B7) (cat# 2280-MC-100) recognizes mouse, but not human Bax protein.

## TREVIGEN®

8405 Helgerman Court, Gaithersburg, MD 20877 USA

Voice: 1-800-TREVIGEN (1-800-873-8443) • 301-216-2800

Fax: 301-560-4973 • e-mail: [info@trevigen.com](mailto:info@trevigen.com) • [www.trevigen.com](http://www.trevigen.com)

### Cell Lysates for Western Blotting:

To prepare total cell lysates, cells are solubilized in 1X SDS gel sample buffer (63 mM Tris, pH 6.8, 10% glycerol, 2% SDS, 2.5% β-mercaptoethanol, and 0.0025% bromophenol blue) at 1 x 10<sup>7</sup> cells per ml. The extracts are heated in a boiling water bath for 5 minutes. Electrophoresis on 4-20% Tris-Glycine SDS-PAGE gels.

### Procedure for Immunoblotting using Peroxidase Detection:

Blotting buffer: 12 mM Tris base, 96 mM Glycine, and 20% MeOH.

Blocking solution: 5% (w/v) nonfat dry milk in TBS, 0.1% Tween®-20.

Antibody solution: 5% (w/v) nonfat dry milk, 0.1% Tween®-20 in TBS.

Transfer the electrophoresed proteins to a nitrocellulose or PVDF membrane and incubate for 1 hour at room temperature in blocking solution. Then, incubate overnight at 4°C in antibody solution containing a 1:1000 dilution of anti-Bax YTH-5B7 antibody. Empirical determination of primary antibody concentration will be required for optimal results.

Wash the membrane at room temperature for 15 minutes with 3 changes of 0.1% Tween®-20 in TBS. Incubate the membrane at room temperature for 1 hour in antibody solution containing anti-mouse conjugated to horseradish peroxidase. Empirical determination of the secondary antibody concentration will be required for optimal results.

Wash the membrane for 15 minutes with 3 changes of 0.1% Tween®-20 in TBS. Develop peroxidase reaction using e.g. chemiluminescence (Trevigen's PeroxyGlow A, cat# 4855-20-13, and PeroxyGlow B, cat# 4855-20-14).

### References:

1. Suzuki, M., R.J. Youle, and N. Tjandra. 2000. Structure of bax. Coregulation of dimer formation and intracellular localization. *Cell* **103**:645-54.
2. Hsu, Y.-T., K.G. Wolter, and R.J. Youle. 1997. Cytosol-to-membrane redistribution of Bax and Bcl-X<sub>L</sub> during apoptosis. *Proc. Natl. Acad. Sci. USA* **94**:3668-3672.
3. Hsu, Y.-T., and R.J. Youle. 1997. Nonionic detergents induce dimerization among members of the Bcl-2 family. *J. Biol. Chem.* **272**:13829-13834.
4. Neuchushtan, A., C.L. Smith, Y.-T. Hsu, and R.J. Youle. 1999. Conformation of the Bax C-terminus regulates subcellular location and cell death. *EMBO J.* **18**:2330-2341.

### Related Products:

Catalog #	Description	Size
4411-PC-100	Anti-Phosphorylated Histone H2AX polyclonal	100 µl
6370-MC-100	Anti-human/murine-Cytochrome C	100 µg
6380-MC-100	Anti-human/murine-Holocytochrome C	100 µg
2290-MC-100	Anti-Bcl-2 mAb (clone YTH 10C4)	100 µg
2291-MC-100	Anti-Bcl-2 mAb (clone YTH 8C8)	100 µg
2282-MC-100	Anti-Bcl-2 mAb (clone YTH 2D2)	100 µg
2281-MC-100	Anti-human-Bax mAb (clone YTH-6A7)	100 µg
2300-MC-100	Anti-Bcl-XL (clone YTH-2H12)	100 µg
6361-PC-100	Anti-human/mouse-PBR polyclonal	100 µl
4335-MC-100	Anti-PAR polymer mAb (10HA)	100 µl
4336-BPC-100	Anti- PAR polymer polyclonal	100 µl
4338-MC-50	Anti-human/murine-PARP mAb (clone C2-10)	50 µg

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 1-800-873-8443