

# TREVIGEN® Product Data

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

## Anti-Bcl-2 YTH-8C8 Monoclonal Antibody

**Catalog #:** 2291-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. The Bcl-2 gene was initially isolated from the t(14;18) chromosomal translocation found in human B-cell follicular lymphomas and was subsequently shown to repress cell death triggered by a diverse array of stimuli. However, the biochemical process by which Bcl-2 regulates cell death is poorly understood.

**Physical State:** This antibody is provided as purified immunoglobulin from mouse ascites in 1X PBS.

**Immunogen:** A synthetic peptide corresponding to amino acids 41 to 54 of the human Bcl-2 sequence.

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

**Specificity:** Anti-Bcl-2 YTH-8C8 reacts with human Bcl-2.

**Storage:** Store at -20 °C. Aliquot to avoid freeze-thaw cycles.

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

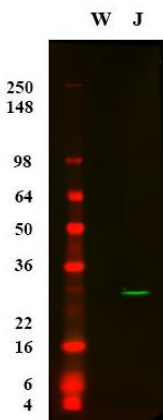


Fig. 1. Western blot analysis of Wehi (W) and Jurkat (J) cell lines. Cells were lysed in Tris-Glycine SDS sample buffer at a concentration of  $1 \times 10^7$  cells/ml and 10 µl of lysates were loaded per well of 4-20% Tris-Glycine gel. Proteins were transferred onto an Immobilon FL membrane and Bcl-2 protein was detected with Trevigen's anti-Bcl-2 (clone YTH-8C8) antibody (cat# 2291-MC-100) and visualized using an IR800-conjugated secondary antibody. The membrane was scanned using an Odyssey Infrared Imaging System (Licor). Trevigen's Bcl-2 antibody (clone YTH-8C8) (cat# 2291-MC-100) recognizes human Bcl-2 protein.

anti-Bcl2 (YTH-8C8) MC (1:1,000)  
+  
Goat anti-Mouse IR800 (1:5,000)

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E03/11/16v3

## 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-Bcl-2 YTH-8C8 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# 4675-096-01, and PeroxyGlow B, cat# 4675-096-02).

**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
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
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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