

TREVIGEN® Product Data

For Research Use Only. Not For Use In Diagnostic Procedures

Anti-Bcl-2 YTH-10C4 Monoclonal Antibody

Catalog #: 2290-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 containing 0.01% sodium azide.

Immunogen: A synthetic peptide corresponding to amino acids 61 to 76 of the mouse Bcl-2 sequence.

Ig Class: IgG₁

Specificity: Anti-Bcl-2 YTH-10C4 cross reacts with mouse and rat Bcl-2.

Storage: Store at 4°C.

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

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 5×10^5 - 1×10^6 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.

Antibody solution: 5% (w/v) nonfat dry milk, 0.05% Tween in TBS.

Transfer the electrophoresed proteins to a nitrocellulose or PVDF membrane and incubate the membrane for 1 hour at room temperature in blocking solution. Incubate the membrane overnight at 4°C in antibody solution containing a 1:1000 dilution of anti-Bcl-2 YTH-10C4 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 0.05% Tween in TBS. Changing the membrane containers often reduces background.

Incubate the membrane at room temperature for 1 hour in antibody solution containing anti-rabbit 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 0.05% Tween in TBS.

Develop peroxidase reaction using e.g. chemiluminescence (Trevigen's PeroxyGlow A, cat# 4855-20-13, and PeroxyGlow B, cat# 4855-20-14).

Related Products:

Catalog #	Description	Size
2291-MC-100	Anti-human-Bcl-2 mAb (clone YTH-8C8)	100 µg
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
2300-MC-100	Anti-Bcl-X _L mAb (clone YTH-2H12)	100 µg
2281-MC-100	Anti-human-Bax mAb (clone YTH-6A7)	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

- References:**
- Suzuki, M., R.J. Youle, and N. Tjandra. 2000. Structure of bax. Coregulation of dimer formation and intracellular localization. *Cell* **103**:645-54.
 - Hsu, Y.-T., K.G. Wolter, and R.J. Youle. 1997. Cytosol-to-membrane redistribution of Bax and Bcl-X_L during apoptosis. *Proc. Natl. Acad. Sci. USA* **94**:3668-3672.
 - 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.
 - 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

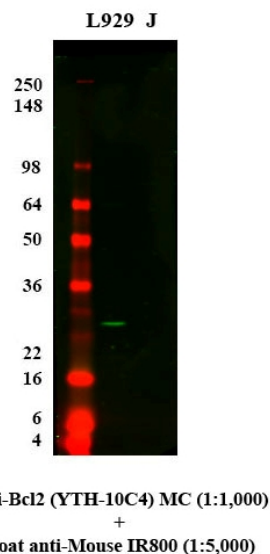


Fig. 1. Western blot analysis of L929 and Jurkat (J) cell lines. Cells were lysed in Tris-Glycine SDS sample buffer at the concentration 10^7 cells/ml and 10 µl of lysates were loaded per well of a 4-20% Tris-Glycine gel. Proteins were transferred onto an Immobilon FL membrane and Bcl-2 protein was detected with Trevigen's anti-Bcl-2 (YTH-10C4) antibody (cat# 2290-MC-100) followed by an IR800-conjugated secondary antibody. The membrane was scanned using an Odyssey Infrared Imaging System (Licor).

TREVIGEN®

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Monoclonal Antibody**

Catalog #: 2290-MC-100

Storage: 4 °C

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