

Technical Data Sheet

PE-CF594 Mouse Anti-Human Bcl-2

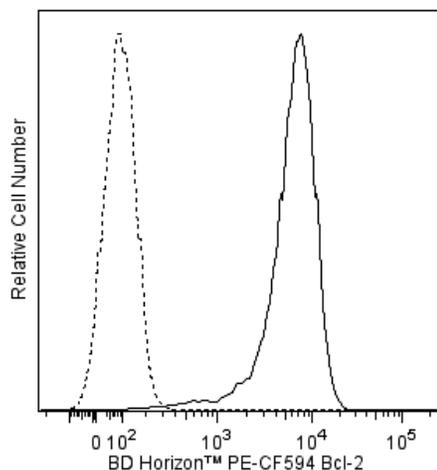
Product Information

Material Number:	563601
Alternate Name:	BCL2; Apoptosis regulator Bcl-2; B-cell CLL/lymphoma 2
Size:	50 tests
Vol. per Test:	5 µl
Clone:	Bcl-2/100
Immunogen:	Human Bcl-2 synthetic peptide aa. 41-54
Isotype:	Mouse (BALB/c) IgG1, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The Bcl-2/100 monoclonal antibody specifically binds to human Bcl-2. It has been reported not to crossreact with mouse Bcl-2. A synthetic peptide corresponding to amino acids 41-54 (GAAPAPGIFSSQPG) of human Bcl-2 was used as the immunogen. This peptide sequence reportedly is not conserved between human and mouse. Programmed cell death (apoptosis) is a normal physiologic process which occurs during embryonic development as well as in maintenance of tissue homeostasis. The apoptotic program is characterized by certain morphological features. These include changes in the plasma membrane such as loss of membrane asymmetry and attachment, a condensation of the cytoplasm and nucleus, and internucleosomal cleavage of DNA. In the final stages, the dying cells become fragmented into “apoptotic bodies” which are rapidly eliminated by phagocytic cells without eliciting significant inflammatory damage to surrounding cells. Members of the Bcl-2 family play a major role in regulating the response of cells to apoptotic signals. Bcl-2 is considered to be novel among proto-oncogenes because it blocks apoptosis in many cell types. Bcl-2 is thought to provide selective survival advantage for cells by blocking apoptosis and thus may contribute to tumorigenesis. Bcl-2 is an approximately 26 kDa intracellular, integral membrane protein found primarily in the nuclear envelope, endoplasmic reticulum and outer mitochondrial membrane.

This antibody is conjugated to BD Horizon™ PE-CF594, which has been developed exclusively by BD Biosciences as a better alternative to PE-Texas Red®. PE-CF594 excites and emits at similar wavelengths to PE-Texas Red® yet exhibits improved brightness and spectral characteristics. Due to PE having maximal absorption peaks at 496 nm and 564 nm, PE-CF594 can be excited by the blue (488-nm), green (532-nm) and yellow-green (561-nm) lasers and can be detected with the same filter set as PE-Texas Red® (eg 610/20-nm filter).



Flow cytometric analysis of Bcl-2 expression in human peripheral blood mononuclear cells. Human PBMC were fixed and permeabilized using BD Cytofix/Cytoperm™ (Cat. No. 554714) followed by staining with either BD Horizon™ PE-CF594 Mouse IgG1, κ Isotype Control (Cat. No. 562292; dashed line histogram) or BD Horizon™ PE-CF594 Mouse Anti-Human Bcl-2 antibody (Cat. No. 563601; solid line histogram). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis was performed using a LSRFortessa™ Cell Analyzer System.

Preparation and Storage

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated with BD Horizon™ PE-CF594 under optimum conditions, and unconjugated antibody and free PE-CF594 were removed.

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
562292	PE-CF594 Mouse IgG1, κ Isotype Control	0.1 mg	X40
554714	BD Cytofix/Cytoperm™ Fixation/Permeablization Kit	250 tests	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100- μ l experimental sample (a test).
2. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
3. An isotype control should be used at the same concentration as the antibody of interest.
4. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
5. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
6. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
8. Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
9. CF™ is a trademark of Biotium, Inc.
10. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
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12. Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using multi-laser cytometers, which may directly excite both PE and CF™594.

References

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- Pezzella F, Jones M, Ralfkiaer E, Ersbøll J, Gatter KC, Mason DY. Evaluation of bcl-2 protein expression and 14;18 translocation as prognostic markers in follicular lymphoma. *Br J Cancer.* 1992; 65(1):87-89. (Clone-specific: Immunohistochemistry)
- Pezzella F, Tse AG, Cordell JL, Pulford KA, Gatter KC, Mason DY. Expression of the bcl-2 oncogene protein is not specific for the 14;18 chromosomal translocation. *Am J Pathol.* 1990; 137(2):225-232. (Immunogen: Immunohistochemistry, Western blot)
- Pezzella F, Turley H, Kuzu I, et al. bcl-2 protein in non-small-cell lung carcinoma. *N Engl J Med.* 1993; 329(10):690-694. (Clone-specific: Immunohistochemistry)
- Reed JC, Meister L, Tanaka S, et al. Differential expression of bcl2 protooncogene in neuroblastoma and other human tumor cell lines of neural origin. *Cancer Res.* 1991; 51(24):6529-6538. (Biology)
- Tanaka S, Saito K, Reed JC. Structure-function analysis of the Bcl-2 oncoprotein. Addition of a heterologous transmembrane domain to portions of the Bcl-2 beta protein restores function as a regulator of cell survival. *J Biol Chem.* 1993; 268(15):10920-10926. (Biology)
- Williams GT. Programmed cell death: apoptosis and oncogenesis. *Cell.* 1991; 65(7):1097-1098. (Biology)

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