

Technical Data Sheet

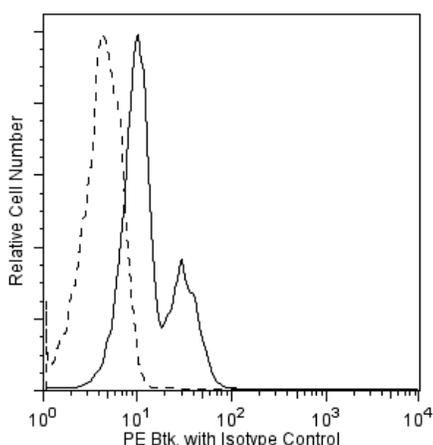
PE Mouse anti-Human Btk

Product Information

Material Number:	558527
Size:	50 tests
Vol. per Test:	20 µl
Clone:	53/BTK
Immunogen:	Human N-Terminal Btk aa. 2-172 Recombinant Protein
Isotype:	Mouse IgG2a
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

Bruton's tyrosine kinase (Btk) is a nonreceptor tyrosine kinase whose function is critical for proper B cell development and signaling. It is a member of the Tec family of kinases which includes Tec and Itk. This family is similar to the src family of tyrosine kinases. However, Tec family members lack the N-terminal myristylation site and the regulatory C-terminal tyrosine that are found in src proteins. In addition to an N-terminal pleckstrin homology (PH) domain, the Tec proteins contain Src homology domains 2 and 3 (SH2 and SH3) and a stretch of 60-80 amino acids between the PH and SH3 domains termed the Tec homology domain. The activity of Btk is regulated by Src-mediated phosphorylation of the kinase domain at tyrosine 551. This event induces Btk kinase activity and subsequent autophosphorylation at tyrosine 223 in the SH3 domain. Phosphorylated Btk then associates with the cell membrane via the interaction of the PH domain with phosphatidylinositol 3, 4, 5-triphosphate. The PH domain is essential for proper activation and function of Btk. A mutation in the PH domain results in Xid, murine X-linked immunodeficiency, and human X-linked agammaglobulinemia.

**Analysis of Btk in human peripheral blood lymphocytes.**

Human whole blood was lysed and fixed with 1X BD Phosflow™ Lyse/Fix Buffer (Cat. No. 558049) for 10-15 minutes at 37°C, then permeabilized (BD Phosflow™ Perm Buffer II, Cat. No. 558052) on ice for 30 minutes, and then stained with either PE Mouse anti-Btk (solid-line histogram) or PE Mouse IgG2a isotype control mAb X39 (Cat. No. 340756, dashed-line histogram). Flow cytometry was performed on a BD FACSCalibur™ flow cytometry system. The figure displays lymphocytes that were selected by their scatter profile, and the cells showing higher Btk expression are B lymphocytes.

Preparation and Storage

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

The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed.

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

Application Notes

Application

Intracellular staining (flow cytometry)	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
558049	Lyse/Fix Buffer 5X	250 ml	(none)
558052	Perm Buffer II	125 ml	(none)

Product Notices

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100-µl experimental sample (a test).
- Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.

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3. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
4. 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.
5. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

References

Mahajan S, Fargnoli J, Burkhardt AL, Kut SA, Saouaf SJ, Bolen JB. Src family protein tyrosine kinases induce autoactivation of Bruton's tyrosine kinase. *Mol Cell Biol.* 1995; 15:5304-5311. (Biology)

Marshall AJ, Niito H, Yun TJ, Clark EA. Regulation of B-cell activation and differentiation by the phosphatidylinositol 3-kinase and phospholipase C γ pathway. *Immunol Rev.* 2000; 176:30-46. (Biology)

Rawlings DJ, Scharenberg AM, Park H, et al. Activation of BTK by a phosphorylation mechanism initiated by SRC family kinases. *Science.* 1996; 271:822-825. (Biology)

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