Technical Data Sheet

PE-CF594 Mouse Anti-Human CD152

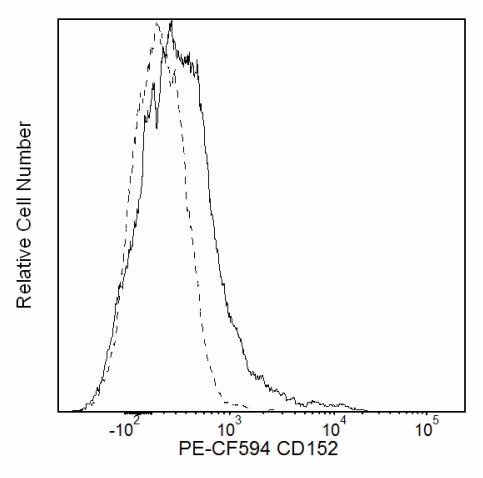
Product Information

Material Number:	**562742**
Alternate Name:	CTLA-4; AILIM; Cytotoxic T-lymphocyte protein 4
Size:	50 tests
Vol. per Test:	5 µl
Clone:	BNI3
Immunogen:	Human CTLA4 Recombinant Protein
Isotype:	Mouse (BALB/c) IgG2a, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The BNI3 monoclonal antibody specifically binds to the human cytolytic T lymphocyte-associated antigen, CTLA-4. CTLA-4 is transiently expressed on activated CD28+ T cells and binds to CD80 and CD86 present on antigen presenting cells (APC) with high avidity. This interaction appears to deliver a negative regulatory signal to the T cell. There are recent reports that indicate that CTLA-4 is also expressed on B cells when cultured with activated T cells, suggesting a possible role of CTLA-4 in the regulation of B-cell response. Immobilized BNI3.1 antibody enhances T-cell proliferation induced by antibody-mediated crosslinking of CD3 and CD28. Recent studies have shown that CD152 can be expressed by regulatory T (Treg) cells. After cellular fixation and permeabilization, the BNI3 antibody can stain intracellular CD152 expressed in T cells including Treg cells. Clone BNI3.1 was studied in the VI Leukocyte Typing Workshop.

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 CD152 expression on concanavalin A-activated peripheral blood mononuclear cells. *Human peripheral blood mononuclear cells were stimulated with concanavalin A for 3 days and then stained with BD Horizon™ PE-CF594 Mouse Anti-Human CD152 antibody (Cat. No. 562742; solid line histogram), or with a BD Horizon™ PE-CF594 Mouse IgG2a, κ Isotype Control (Cat. No. 562306; dashed line histogram). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable activated cells. Flow cytometry was performed using a BD™ LSR II Flow Cytometer 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

Flow cytometry	Routinely Tested

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Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 ml	(none)
562306	PE-CF594 Mouse IgG2a, κ Isotype Control	0.1 mg	G155-178

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-µl 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/pharmingen/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.
11. This product is provided under an Agreement between BIOTIUM and BD Biosciences. The manufacture, use, sale, offer for sale, or import of this product is subject to one or more patents or pending applications owned or licensed by Biotium, Inc. This product, and only in the amount purchased by buyer, may be used solely for buyer's own internal research, in a manner consistent with the accompanying product literature. No other right to use, sell or otherwise transfer (a) this product, or (b) its components is hereby granted expressly, by implication or by estoppel. This product is for research use only. Diagnostic uses require a separate license from Biotium, Inc. For information on purchasing a license to this product including for purposes other than research, contact Biotium, Inc., 3159 Corporate Place, Hayward, CA 94545, Tel: (510) 265-1027. Fax: (510) 265-1352. Email: btinfo@biotium.com.
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

Castan J, Klauenberg U, Kalmar P, Fleischer B, Broker BM. Expression of CTLA-4 (CD152) on human medullary CD4+ thymocytes. *Med Microbiol Immunol (Berl).* 1998; 187(1):49-52. (Immunogen: Immunofluorescenc)

Castan J, Tenner-Racz K, Racz P, Fleischer B, Broker BM. Accumulation of CTLA-4 expressing T lymphocytes in the germinal centres of human lymphoid tissues. *Immunology.* 1997; 90(2):265-271. (Immunogen: ELISA, Immunofluorescenc)

Lindsten T, Lee KP, Harris ES, et al. Characterization of CTLA-4 structure and expression on human T cells. *J Immunol.* 1993; 151(7):3489-3499. (Biology)

Morton PA, Fu XT, Stewart JA, et al. Differential effects of CTLA-4 substitutions on the binding of human CD80 (B7-1) and CD86 (B7-2). *J Immunol.* 1996; 156(3):1047-1054. (Biology)

Wang H, Shih CC, Waters JB, Balderas RS, Rosenberg J, Huang EC-M, Chen Z. CD152 (CTLA4) Workshop: Expression and function of CD152 on human T cells: A study using a mouse anti-human CD152 monoclonal antibody BNI3.1. In: Kishimoto T, von dem Borne AEG, Goyert SM,et al., ed. *Leucocyte Typing VI: White Cell Differentiation Antigens.* London: Garland Publishing; 1997:97-98. (Clone-specific: Blocking, (Co)-stimulation, Flow cytometry, Functional assay, Inhibitio)

Zola H, Swart B, Nicholson I, Voss E. *Leukocyte and Stromal Cell Molecules. The CD Markers.* Hoboken, New Jersey: John Wiley & Sons, Inc.; 2007:1-581. (Biology)

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