Technical Data Sheet

PE-CF594 Rat Anti-Mouse Foxp3

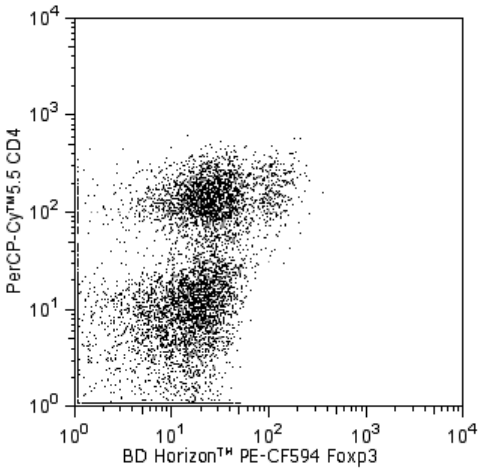
Product Information

Material Number:	**562466**
Alternate Name:	Forkhead box p3; Forkhead box protein p3; JM2; Scurfin; Scurfy; Sf
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	MF23
Immunogen:	Mouse Foxp3 Recombinant Protein
Isotype:	Rat IgG2b
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The MF23 monoclonal antibody specifically binds to mouse Foxp3. Foxp3 is a 50-55 kDa protein also known as Forkhead box p3, JM2, or IPEX. It is a member of the forkhead or winged helix family of transcription factors and is specifically expressed by T regulatory (Treg) cells. Foxp3 has been reported to be a key regulatory protein for Treg cell development and function. Ectopic expression of Foxp3 in conventional T cells is sufficient to induce suppressive activity, repress the production of cytokines such as IL2 and IFN-γ, and upregulate Treg cell-associated molecules such as CD25, CTLA4 and GITR. It has been found that the mutation of Foxp3 is responsible for "scurfy" mice. When overexpressed, Foxp3 leads to poor T cell proliferation and activation. Immunoblotting with MF23 antibody has confirmed it recognizes an epitope between 1-87 amino acids in the N-terminal domain.

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).



Multicolor flow cytometric analysis of Foxp3 expression in BALB/c mouse splenocytes. *Splenocytes from BALB/c mice were fixed and permeabilized using working solutions of Mouse Foxp3 Buffers (Mouse Foxp3 Buffer Set, Cat. No. 560409). The cells were then stained with PerCP-Cy™5.5 Rat anti-Mouse CD4 (Cat. No. 550954/561115) and BD Horizon™ PE-CF594 Rat anti-Mouse Foxp3 antibodies (Cat. No. 562466). The flow cytometric dot plot shows the correlated expression of Foxp3 versus CD4 for gated events with the forward and side light-scattering characteristics of intact lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometry 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

BD Biosciences

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Suggested Companion Products

Catalog Number	Name	Size	Clone
560409	Mouse Foxp3 Buffer Set	100 Tests	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
550954	PerCP-Cy™5.5 Rat Anti-Mouse CD4	0.1 mg	RM4-5
561115	PerCP-Cy™5.5 Rat Anti-Mouse CD4	25 µg	RM4-5
562308	PE-CF594 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1
554657	Stain Buffer (BSA)	500 mL	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
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. 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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
8. CF™ is a trademark of Biotium, Inc.
9. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
10. 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.
11. 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.
12. Cy is a trademark of GE Healthcare.
13. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Hori S, Nomura T, Sakaguchi S. Control of regulatory T cell development by the transcription factor Foxp3. *Science.* 2003; 299(5609):1057-1061. (Biology)

Jinushi M, Sato M, Kanamoto A, et al. Milk fat globule epidermal growth factor-8 blockade triggers tumor destruction through coordinated cell-autonomous and immune-mediated mechanisms. *J Exp Med.* 2009; 206(6):1317-1326. (Clone-specific: Flow cytometry)

Ono M, Yaguchi H, Ohkura N, et al. Foxp3 controls regulatory T-cell function by interacting with AML1/Runx1. *Nature.* 2007; 446(7136):685-689. (Biology)

Vasconcellos R, Carter NA, Rosser EC, Mauri C. IL-12p35 subunit contributes to autoimmunity by limiting IL-27-driven regulatory responses. *J Immunol.* 2011; 187(6):3402-3412. (Clone-specific: Flow cytometry)

Zheng Y, Rudensky AY. Foxp3 in control of the regulatory T cell lineage. *Nat Immunol.* 2007; 8:457-462. (Biology)