# **Technical Data Sheet**

# PE-CF594 Rat Anti-Human GM-CSF

#### **Product Information**

Material Number: 562857

Alternate Name: CSF2; Colony stimulating factor 2 (granulocyte-macrophage); CSF; GMCSF

 Size:
 50 tes

 Vol. per Test:
 5 μl

Clone: BVD2-21C11

Immunogen: Recombinant human GM-CSF

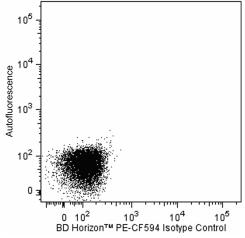
Isotype:Rat (LEW) IgG2aReactivity:QC Testing: Human

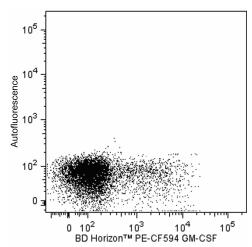
**Storage Buffer:** Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The BVD2-21C11 monoclonal antibody specifically binds to human Granulocyte/Macrophage - Colony Stimulating Factor (GM-CSF). Human GM-CSF is encoded by the *CSF2* gene and is also known as Colony Stimulating Factor 2. GM-CSF is produced by activated T lymphocytes, macrophages, endothelial cells, fibroblasts, stromal cells and other cell types including B lymphocytes, mast cells, eosinophils, and osteoblasts. GM-CSF stimulates the survival, proliferation and/or differentiation of various cell types including neutrophils, eosinophils, macrophages, dendritic cells, megakaryocytes, erythroid cells, endothelial cells and their precursors. The immunogen used to generate the BVD2-21C11 hybridoma was recombinant human GM-CSF. The BVD2-21C11 antibody has been reported to crossreact with GM-CSF from the rhesus monkey. BVD2-21C11 is a neutralizing antibody.

This antibody is conjugated to BD Horizon<sup>TM</sup> 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).





Multiparameter flow cytometric analysis of GM-CSF expressed in stimulated human peripheral blood mononuclear cells. HiCK-1 (Human intracellular CytoKine-1) Cytokine Positive Control Cells (Cat. No. 555061) were permeabilized with BD Perm/Wash™ Buffer (Cat. No. 554723). The cells were then stained with either BD Horizon™ PE-CF594 Rat IgG2a, k Isotype Control (Cat No. 562302, Left Panel) or BD Horizon™ PE-CF594 Rat Anti-Human GM-CSF antibody (Cat No. 562857, Right Panel) by using BD Biosciences Intracellular Cytokine Staining protocol. Two-color flow cytometric dot plots showing the expressed levels of GM-CSF (or Ig isotype control staining) versus forward light scatter were derived from gated events with the forward and side light-scatter characteristics of intact lymphocytes. Flow cytometric analysis 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.

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## **Application Notes**

#### Application

Untracellular staining (flow cytometry)	Routinely Lested	

## **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
554656	Stain Buffer (FBS)	500 ml	(none)	
562302	PE-CF594 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95	
555061	HiCK-1 Human Cytokine Positive Control Cells	1.0 ml	(none)	
554723	Perm/Wash Buffer	100 ml	(none)	

#### **Product Notices**

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> 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.
- 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.
- CF<sup>TM</sup> 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 CFTM594.

### References

Abrams J. Immunoenzymetric assay of mouse and human cytokines using NIP-labeled anti-cytokine antibodies. Curr Protoc Immunol. 2001; 1:6.20-6.21. (Clone-specific: ELISA)

Abrams JS, Roncarolo MG, Yssel H, Andersson U, Gleich GJ, Silver JE. Strategies of anti-cytokine monoclonal antibody development: immunoassay of IL-10 and IL-5 in clinical samples. *Immunol Rev.* 1992; 127:5-24. (Clone-specific: ELISA, Flow cytometry, Neutralization)

Abrams JS, Silver JE, Van Dyke RE, Gleich GI. Eosinophil-active cytokines in human disease: development and use of monoclonal antibodies to IL-3, IL-5 and GMCSF. In: Gleich GJ and Kay AB, ed. *Eosinophils in Allergy and Inflammation*. New York: Dekker; 1994:133-157. (Immunogen: ELISA, Neutralization) Bacchetta R, de Waal Malefijt R, Yssel H. Host-reactive CD4+ and CD8+ T cell clones isolated from a human chimera produce IL-5, IL-2, IFN-gamma and granulocyte/macrophage-colony-stimulating factor but not IL-4. *J Immunol*. 1990; 144(3):902-908. (Clone-specific: ELISA, Neutralization) Gasson JC. Molecular physiology of granulocyte-macrophage colony-stimulating factor. *Blood*. 1991; 77(6):1131-1145. (Biology)

Kita H, Ohnishi T, Okubo Y, Weiler D, Abrams JS, Gleich GJ. Granulocyte/macrophage colony-stimulating factor and interleukin 3 release from human peripheral blood eosinophils and neutrophils. *J Exp Med.* 1991; 174(3):745-748. (Clone-specific: ELISA, Neutralization)

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. *J Immunol Methods*. 1995; 188(1):117-128. (Methodology: Flow cytometry)

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