

## Technical Data Sheet

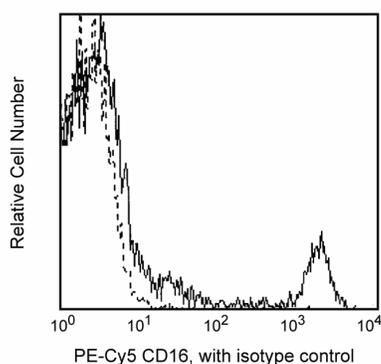
## PE-Cy™5 Mouse Anti-Human CD16

## Product Information

<b>Material Number:</b>	561725
<b>Alternate Name:</b>	FcRIII; Fc-gamma RIII; FCG3; FCGR3; FCGRIII; FcγRIII; IGFR3
<b>Size:</b>	25 tests
<b>Vol. per Test:</b>	20 μl
<b>Clone:</b>	3G8
<b>Immunogen:</b>	Human polymorphonuclear leukocytes
<b>Isotype:</b>	Mouse IgG1, κ
<b>Reactivity:</b>	QC Testing: Human Reported: Baboon, Rhesus, Cynomolgus
<b>Workshop:</b>	IV N409
<b>Storage Buffer:</b>	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

## Description

The 3G8 monoclonal antibody specifically binds to the 50-65 kDa transmembrane form of the IgG Fc Receptor (FcγRIII), a human NK-cell-associated antigen. CD16 is expressed on NK cells as well as macrophages and granulocytes. Reports indicate that CD16 plays a role in signal transduction and NK cell activation. The 3G8 antibody blocks the binding of soluble immune complexes to granulocytes.



Profile of peripheral blood lymphocytes analyzed on a BD FACScan™ (BDIS, San Jose, CA)

## Preparation and Storage

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

The antibody was conjugated with PE-Cy5 (formerly known as BD Cy-Chrome™) under optimum conditions, and unconjugated antibody and free PE-Cy5 were removed.

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

## Application Notes

## Application

Flow cytometry	Routinely Tested
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## Suggested Companion Products

Catalog Number	Name	Size	Clone
555750	PE-Cy™5 Mouse IgG1 κ Isotype Control	100 tests	MOPC-21
554656	Stain Buffer (FBS)	500 ml	(none)

## Product Notices

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100-μl experimental sample (a test).
- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
- Source of all serum proteins is from USDA inspected abattoirs located in the United States.

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5. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
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7. 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.
8. 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.
9. PE-Cy5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the PE-Cy5 tandem fluorochrome, extra care must be taken when using dual-laser cytometers which may directly excite both PE and Cy5™.
10. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
11. PE-Cy5 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by the 488 nm light of an Argon ion laser and serves as an energy donor, coupled to the cyanine dye Cy5, which acts as an energy acceptor and fluoresces at 670 nm. BD Biosciences Pharmingen has maximized the fluorochrome energy transfer in PE-Cy5, thus maximizing its fluorescence emission intensity, minimizing residual emission from PE, and minimizing lot-to-lot variation.
12. PE-Cy5 tandem fluorochromes have been reported to bind some classes of human macrophages and granulocytes via Fc receptors, and PE has been reported to bind to mouse B lymphocytes via Fc receptors. Preincubation of mouse leukocytes with Mouse BD Fc Block™ purified anti-mouse CD16/CD32 mAb 2.4G2 can reduce the non-specific binding of PE-Cy5-conjugated reagents to mouse B cells. However, PE-Cy5 conjugated reagents should not be used to stain splenocytes of SJL, NOD, and MRL mice as B lymphocytes and/or other leukocytes have been reported to non-specifically stain regardless of the use of Mouse BD Fc Block™ (the CD72c complex has been implicated for PE-Cy5 binding in these strains). Reagents conjugated to PE, PerCP, PerCP-Cy5.5, APC, and APC-Cy7 tandem fluorochrome can be used on leukocytes from these mouse strains.
13. An isotype control should be used at the same concentration as the antibody of interest.

## References

- Barclay NA, Brown MH, Birkeland ML, et al, ed. *The Leukocyte Antigen FactsBook*. San Diego, CA: Academic Press; 1997. (Biology)
- Fleit HB, Wright SD, Unkeless JC. Human neutrophil Fc gamma receptor distribution and structure. *Proc Natl Acad Sci U S A*. 1982; 79(10):3275-3279. (Biology)
- Knapp W, Dorken B, Rieber EP, et al, ed. *Leucocyte Typing IV*. New York: Oxford University Press; 1989:1-1208. (Clone-specific)
- Stroncek DF, Skubitz KM, Plachta LB, et al. Alloimmune neonatal neutropenia due to an antibody to the neutrophil Fc-gamma receptor III with maternal deficiency of CD16 antigen. *Blood*. 1991; 77(7):1572-1580. (Biology)
- Wirthmueller U, Kurosaki T, Murakami MS, Ravetch JV.. Signal transduction by Fc gamma RIII (CD16) is mediated through the gamma chain. *J Exp Med*. 1992; 175(5):1381-1390. (Biology)