

## Technical Data Sheet

**BV786 Mouse Anti-Human CD16****Product Information**

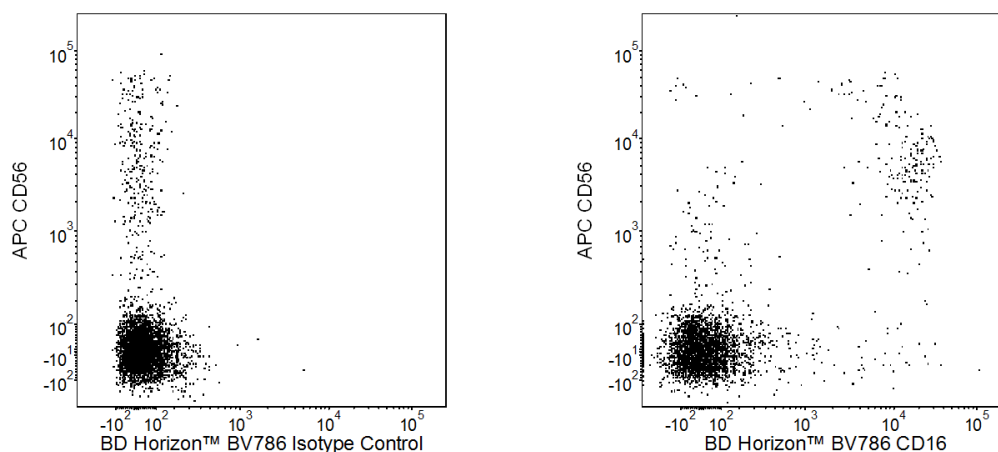
<b>Material Number:</b>	<b>563690</b>
<b>Alternate Name:</b>	FcRIII; Fc-gamma RIII; FCG3; FCGR3; FCGRIII; FcγRIII; IGFR3
<b>Size:</b>	100 Tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	3G8
<b>Immunogen:</b>	Human polymorphonuclear leukocytes
<b>Isotype:</b>	Mouse (BALB/c x DBA/2) IgG1, κ
<b>Reactivity:</b>	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon
<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. The 3G8 antibody is reported (Vosseveld *et al.*, 1997) to increase intracellular calcium levels in human neutrophils by interacting with both FcγRIIa and FcγRIIb molecules. This antibody has also been reported to induce homotypic neutrophil aggregation.

This clone also cross-reacts with a subset of peripheral blood lymphocytes and monocytes, but not granulocytes, of baboon and both rhesus and cynomolgus macaque monkeys. Multi-color analysis reveals that the distribution on lymphocytes is similar to that found in human studies with the majority of CD16-positive lymphocytes being both CD3 and CD20 negative.

The antibody was conjugated to BD Horizon™ BV786 which is part of the BD Horizon Brilliant™ Violet family of dyes. This dye is a tandem fluorochrome of BD Horizon™ BV421 with an Ex Max of 405-nm and an acceptor dye with an Em Max at 786-nm. BD Horizon™ BV786 can be excited by the violet laser and detected in a filter used to detect Cy7™-like dyes (eg, 780/60-nm filter).



**Two-color flow cytometric analysis of human CD16 expression on human peripheral blood cells.** Human whole blood was stained with APC Mouse Anti-Human CD56 antibody (Cat. No. 555518) and either BD Horizon™ BV786 Mouse IgG1, κ Isotype Control (Cat. No. 563330; Left Panel) or BD Horizon™ BV786 Mouse Anti-Human CD16 antibody (Cat. No. 563689/563690; Right Panel). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The two-color flow cytometric dot plots show the correlated expression patterns of CD16 (or Ig Isotype control staining) versus CD56 for gated events with the forward and side light-scatter characteristics of viable peripheral blood lymphocytes. Flow cytometric analysis was performed using a BD™ LSR II Flow Cytometer System.

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## 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™ BV786 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV786 were removed.

## Application Notes

### Application

Flow cytometry

Routinely Tested

### Recommended Assay Procedure:

For optimal and reproducible results, BD Horizon Brilliant Stain Buffer should be used anytime two or more BD Horizon Brilliant dyes are used in the same experiment. Fluorescent dye interactions may cause staining artifacts which may affect data interpretation. The BD Horizon Brilliant Stain Buffer was designed to minimize these interactions. More information can be found in the Technical Data Sheet of the BD Horizon Brilliant Stain Buffer (Cat. No. 563794).

### Suggested Companion Products

Catalog Number	Name	Size	Clone
563689	BV786 Mouse Anti-Human CD16	25 Tests	3G8
555518	APC Mouse Anti-Human CD56	100 Tests	B159
555899	Lysing Buffer	100 mL	(none)
349202	BD FACS™ Lysing Solution	100 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
563330	BV786 Mouse IgG1, k Isotype Control	50 µg	X40
554657	Stain Buffer (BSA)	500 mL	(none)
563794	Brilliant Stain Buffer	100 Tests	(none)

### Product Notices

1. 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).
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. 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. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [wwwbdbiosciences.com/colors](http://wwwbdbiosciences.com/colors).
6. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
7. Cy is a trademark of GE Healthcare.
8. BD Horizon Brilliant Violet 786 is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,227,187; 8,455,613; 8,575,303; 8,354,239.
9. BD Horizon Brilliant Stain Buffer is covered by one or more of the following US patents: 8,110,673; 8,158,444; 8,575,303; 8,354,239.
10. Please refer to [wwwbdbiosciences.com/pharmingen/protocols](http://wwwbdbiosciences.com/pharmingen/protocols) for technical protocols.

### References

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- Perussia B, Starr S, Abraham S, Fanning V, Trinchieri G. Human natural killer cells analyzed by B73.1, a monoclonal antibody blocking Fc receptor functions. I. Characterization of the lymphocyte subset reactive with B73.1. *J Immunol*. 1983; 130(5):2133-2141. (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. (Clone-specific: Immunofluorescence, Immunoprecipitation)
- Vossebeld PJ, Homburg CH, Roos D, Verhoeven AJ. The anti-Fc gamma RIII mAb 3G8 induces neutrophil activation via a cooperative action of Fc gamma RIIIb and Fc gamma RIIa. *Int J Biochem Cell Biol*. 1997; 29(3):465-473. (Clone-specific: Activation, Bioassay, Calcium Flux)
- Wirhmueller 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. (Clone-specific: Activation, Bioassay, Calcium Flux, Cell separation, Flow cytometry, Immunoprecipitation)
- Zola H. *Leukocyte and stromal cell molecules : the CD markers*. Hoboken, N.J.: Wiley-Liss; 2007:1-581. (Biology)

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563690 Rev. 2

