Technical Data Sheet

# BV510 Mouse Anti-Human CD16

## Product Information

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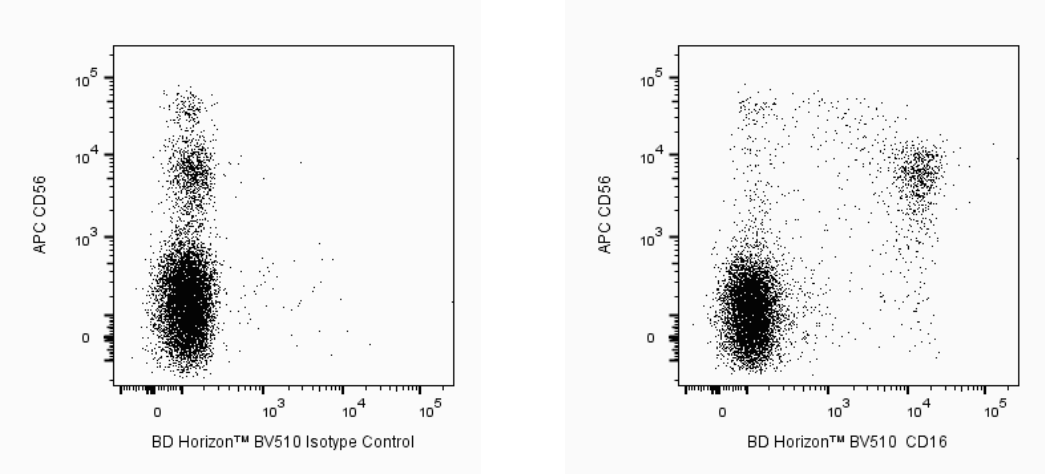
| **Material Number:** | **563829** |
| **Alternate Name:** | CD16;CD16A;FCGR3A;FcγRIIIA;FcRIIIa;CD16B;FCGR3B;FcγRIIIB;FcRIIIb |
| **Size:** | 25 Tests |
| **Vol. per Test:** | 5 µl |
| **Clone:** | 3G8 |
| **Immunogen:** | Human polymorphonuclear leukocytes |
| **Isotype:** | Mouse (BALB/c x DBA/2) IgG1, κ |
| **Reactivity:** | QC Testing: Human<br>Tested in Development: Rhesus, Cynomolgus, Baboon |
| **RRID:** | AB_2744296 |
| **Workshop:** | IV N409; V MR5, NK80 |
| **Storage Buffer:** | Aqueous buffered solution containing BSA and ≤0.09% sodium azide. |

## Description

The 3G8 monoclonal antibody specifically recognizes CD16a and CD16b, low affinity receptors for the Fc region of IgG. CD16a is ~50-65 kDa type I transmembrane glycoprotein that is encoded by *FCGR3A* (Fc fragment of IgG receptor IIIa) which belongs to the immunoglobulin superfamily. CD16a is also known as Fc-gamma RIII-alpha (Fc-gamma RIIIa or FcγRIIIA) or FcRIIIa and is expressed on natural killer cells, activated monocytes, macrophages, γδ T cells, immature thymocytes, and mast cells. CD16a binds immune-complexed or aggregated IgG and associates with CD247/TCRζ in NK cells and FcεRIγ chains in phagocytes and mast cells to transduce intracellular signals. CD16a functions in antibody-dependent cellular cytotoxicity (ADCC) and other antibody-dependent responses including phagocytosis, cytokine production or mediator release. CD16b is a ~48 kDa glycophosyl-phosphatidylinositol (GPI)-linked form that is encoded by *FCGR3B* (Fc fragment of IgG receptor IIIb). CD16b is also known as Fc-gamma RIII-beta (Fc-gamma RIIIb or FcγRIIIB) or FcRIIIb and is expressed on neutrophils and activated eosinophils. The extracellular region of CD16b is highly homologous to CD16a. CD16b also serves as a receptor for the Fc region of IgG and can bind immune-complexed or aggregated IgG and may be involved in neutrophil adhesion.

The 3G8 antibody also crossreacts with a subset of peripheral blood lymphocytes and monocytes, but not granulocytes, of baboon, rhesus, and cynomolgus monkeys. Multicolor 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™ BV510 which is part of the BD Horizon Brilliant™ Violet family of dyes. With an Ex Max of 405-nm and Em Max at 510-nm, BD Horizon™ BV510 can be excited by the violet laser and detected in the BD Horizon™ V500 (525/50-nm) filter set. BD Horizon™ BV510 conjugates are useful for the detection of dim markers off the violet laser.




***Two-color flow cytometric analysis of human CD16 expression on human peripheral blood cells.*** *Human peripheral blood cells were stained with APC Mouse Anti-Human CD56 antibody (Cat. No. 555518) and either BD Horizon™ BV510 Mouse IgG1, κ Isotype Control (Cat. No. 562946; Left Panel) or BD Horizon™ BV510 Mouse Anti-Human CD16 antibody (Cat. No. 563829/563830; Right Panel). The erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). The two-color flow cytometric dot plots show the correlated expression patterns of CD16 versus CD56 (or Ig Isotype control staining) for gated events with the forward and side light-scatter characteristics of intact 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™ BV510 under optimum conditions, and unconjugated antibody and free BD Horizon™ BV510 were removed.

## Application Notes

**Application**

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| 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. 563794/566349) or the BD Horizon Brilliant Stain Buffer Plus (Cat. No. 566385).

## Suggested Companion Products

| Catalog Number | Name | Size | Clone |
|---|---|---|---|
| 554656 | Stain Buffer (FBS) | 500 mL | (none) |
| 566349 | Brilliant Stain Buffer | 1000 Tests | (none) |
| 566385 | Brilliant Stain Buffer Plus | 1000 Tests | (none) |
| 563794 | Brilliant Stain Buffer | 100 Tests | (none) |
| 349202 | Lysing Solution 10X Concentrate | 100 mL | (none) |
| 555899 | Lysing Buffer | 100 mL | (none) |
| 562946 | BV510 Mouse IgG1, k Isotype Control | 50 µg | X40 |
| 563830 | BV510 Mouse Anti-Human CD16 | 100 Tests | 3G8 |
| 555518 | APC Mouse Anti-Human CD56 (NCAM-1) | 100 Tests | B159 |

## 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. 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. Species cross-reactivity detected in product development may not have been confirmed on every format and/or application.
5. 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.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. BD Horizon Brilliant Violet 510 is covered by one or more of the following US patents: 8,575,303; 8,354,239.
8. 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.
9. Please refer to http://regdocs.bd.com to access safety data sheets (SDS).
10. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.


Knapp W. W. Knapp .. et al., ed. *Leucocyte typing IV : white cell differentiation antigens.* Oxford New York: Oxford University Press; :1-1182. (Clone-specific)

Zola H. *Leukocyte and stromal cell molecules : the CD markers.* Hoboken, N.J.: Wiley-Liss; (Biology)

Fleit HB, Wright SD, Unkeless JC. Human neutrophil Fc gamma receptor distribution and structure. *Proc Natl Acad Sci U S A.* 1982; :3275-3279. (Immunogen: Blocking, Immunoprecipitation, Inhibition, Radioimmunoassay)

Perussia B, Trinchieri G, Jackson A, et al. The Fc receptor for IgG on human natural killer cells: phenotypic, functional, and comparative studies with monoclonal antibodies. *J Immunol.* 1984; :180-189. (Clone-specific: Flow cytometry, Functional assay, Inhibition)

Schmidt RE. Non-lineage/natural killer section report: new and previously defined clusters. In: Knapp W. W. Knapp .. et al., ed. *Leucocyte typing IV : white cell differentiation antigens.* Oxford New York: Oxford University Press; :517-542. (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; :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 actin of Fc gamma RIIIb and Fc gamma RIIa. *Int J Biochem Cell Biol.* 1997; :465-473. (Clone-specific: Activation, Functional assay)

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; :1381-1390. (Clone-specific: Activation, Functional assay, Immunoprecipitation)