Technical Data Sheet

PE-Cy™7 Mouse Anti-Human CD16

Product Information

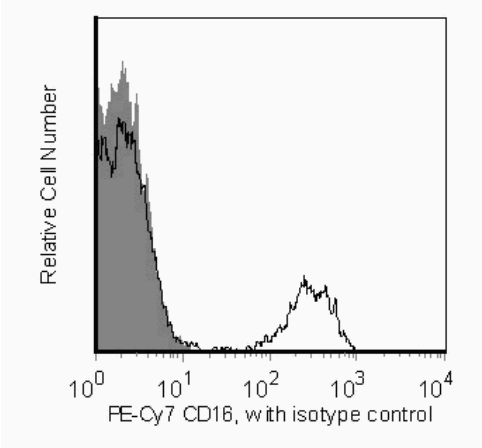
Material Number:	**560716**
Alternate Name:	CD16;CD16A;FCGR3A;FcγRIIIA;FcRIIIa;CD16B;FCGR3B;FcγRIIIB;FcRIIIb
Size:	50 Tests
Vol. per Test:	5 µl
Clone:	3G8
Immunogen:	Human polymorphonuclear leukocytes
Isotype:	Mouse (BALB/c x DBA/2) IgG1, κ
Reactivity:	QC Testing: Rhesus, Cynomolgus, Baboon Tested in Development: Human
RRID:	AB_1727433
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.

This clone also cross-reacts with a subset of peripheral blood lymphocytes and monocytes, but not granulocytes, of baboon and both rhesus and cynomologus 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.



Flow cytometric analysis for CD16 in Rhesus macaque peripheral blood mononuclear cells (PBMC). *PBMC from Rhesus macaque were stained with either a PE-Cy™7 Mouse IgG1, κ isotype control (Cat. No.557872; shaded) or with the PE-Cy™7 Mouse Anti-Human CD16 antibody (Cat. No. 560716/560918/557744; unshaded). Histograms were derived from gated events based on light scattering characteristics for lymphocytes. Flow cytometry was performed on a BD™ LSR II flow cytometry 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 PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)
555899	Lysing Buffer	100 mL	(none)
349202	Lysing Solution 10X Concentrate	100 mL	(none)
557872	PE-Cy™7 Mouse IgG1 κ Isotype Control	100 Tests	MOPC-21
560918	PE-Cy™7 Mouse Anti-Human CD16	25 Tests	3G8
557744	PE-Cy™7 Mouse Anti-Human CD16	100 Tests	3G8

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. 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. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD™ Stabilizing Fixative (Cat. No. 338036).
6. 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.
7. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
9. Cy is a trademark of GE Healthcare.
10. Species cross-reactivity detected in product development may not have been confirmed on every format and/or application.
11. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.

References

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