Technical Data Sheet

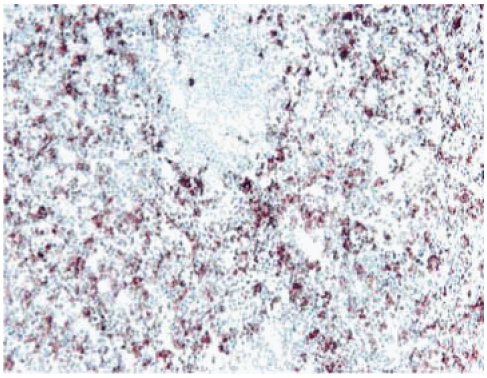
Purified Mouse Anti-Human CD16

Product Information

Material Number:	**550383**
Alternate Name:	FcRIII; Fc-gamma RIII; FCG3; FCGR3; FCGRIII; FcγRIII; IGFR3
Size:	1.0 ml
Concentration:	125 µg/ml
Clone:	3G8
Immunogen:	Human polymorphonuclear leukocytes
Isotype:	Mouse (BALB/c x DBA/2) IgG1, κ
Reactivity:	QC Testing: Human Reported Reactivity: Rhesus, Cynomolgus, Baboon
Workshop:	IV N409
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, 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 (Vossebeld *et al*., 1997) to increase intracellular calcium levels in human neutrophils by interacting with both FcγRIIa and FcγRIIIb molecules. This antibody has also been reported to induce homotypic neutrophil aggregation.



Immunohistochemical staining of human CD16 positive cells. *Frozen sections of normal human spleen was stained with Purified Mouse Anti-Human CD16 (Cat. No. 550383) antibody. Cells positive for CD16 can be identified by intense brown labeling of their cell membranes. Amplification 20X.*

Preparation and Storage

Store undiluted at 4°C.

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

Application Notes

Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunohistochemistry-zinc-fixed	Tested During Development
Immunohistochemistry-paraffin	Not Recommended

Recommended Assay Procedure:

Immunohistochemistry: The 3G8 clone specific for human CD16 is recommended to test for immunohistochemical staining of acetone-fixed frozen sections. Tissue tested was human spleen and tonsil. The antibody stains NK cells and also macrophages and granulocytes. The isotype control recommended for use with this antibody is purified mouse IgG1 (Cat. No. 550878). For optimal indirect immunohistochemical staining, the 3G8 antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with polyclonal, biotin conjugated anti-mouse Igs (multiple adsorbed) (Cat. No. 550337) as the secondary antibody and Streptravidin-HRP (Cat. No. 550946) together with the DAB detection system (Cat. No. 550880). The clone 3G8 is not recommended for formalin-fixed paraffin embedded sections.

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Suggested Companion Products

Catalog Number	Name	Size	Clone
550337	Biotin Goat Anti-Mouse Ig (Multiple Adsorption)	1.0 ml	Polyclonal
550946	Streptavidin HRP	50 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)
550878	Purified Mouse IgG1 κ Isotype Control	1.0 ml	MOPC-31C
559148	Antibody Diluent for IHC	125 ml	(none)
551011	Anti-Mouse Ig HRP Detection Kit	200 tests	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. This antibody has been developed for the immunohistochemistry application. However, a routine immunohistochemistry test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
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. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
6. An isotype control should be used at the same concentration as the antibody of interest.
7. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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, Dörken B, Gilks WR, et al, ed. *Leucocyte Typing IV.* New York, NY: Oxford University Press; 1989:1-1182. (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)

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; 29(3):465-473. (Clone-specific)

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)

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