



	CLONE 1	CLONE 2
Specificity	CD64	CD16
Clone	022CL-3	3G8
Hybridoma	P3/NS/1-Ag4-1 x BALB/c	SP2/0 x BALB/c
Immunogen	Human monocytes	Human neutrophils
Ig Chain	IgG1	IgG1
Species	Mouse	Mouse
Source	Conditional media	Conditional media
Purification	Affinity chromatography	Affinity chromatography
Fluorescence	Excites 486-580 nm / Emits 568-590 nm	Excites 486-580 nm / Emits 710-800 nm
Conjugation	PE (Phycoerythrin)	PC7 (Phycoerythrin-cyanine 7)
Molar Ratio	PE/Protein: 0.5-1.5	PC7/Protein: 0.5-1.5

**For Research Use Only.
Not for use in diagnostic procedures**

SPECIFICITY

CD64 (FcγRI) and CD16 (FcγRIII) along with CD32 (FcγRII) are three structurally similar, yet distinct, receptors for IgG found on human leukocytes.¹ The association of CD64, CD16, and CD32 with the Fc receptor γ chain homodimer is required for its signal transduction activity (γ chain is also a subunit of FcεRI, FcγRIIIA and FcαR)^{2,3} All three Fcγ receptors bind polymeric or aggregated IgG, however CD64 is the only one with high enough affinity to be able to bind monomeric IgG.^{1,4} Both CD64 and CD16 are involved in antibody-dependent cell-mediated cytotoxicity (ADCC), clearance of immune complexes, and phagocytosis of IgG opsonized targets.^{5,6}

CD64 antigen is a single chain, heavily N-glycosylated type I transmembrane molecule with a molecular weight of 72 kDa. CD64 is constitutively expressed on monocytes, macrophages, and a subset of dendritic cells.^{5,6} The expression of CD64 on polymorphonuclear neutrophils is weak but can be upregulated by interferon-γ or granulocyte colony-stimulating factor.⁷

CD16 exists in two different forms encoded by two separate genes: FcγRIIIA and FcγRIIIB, resulting in alternative membrane-anchored molecules. The FcγRIIIA is a transmembrane form (50-65 kDa) expressed on NK cells, monocytes and macrophages. The FcγRIIIB form is a glycoposphatidylinositol (GPI)-anchored receptor that is expressed only on neutrophils.⁸ The CD16 antigen can be non-covalently associated within the membrane of NK cells to the CD3ζ chain, or to the FcεR1γ chain.^{9,10}

The 22CL-3 monoclonal antibody and the 3G8 monoclonal antibody were assigned to their respective cluster of differentiations at the 4th International Workshop on Human Leukocyte Differentiation Antigens in Vienna, Austria, in 1989.¹¹

REAGENTS

IOTest CD64-PE/CD16-PC7 Conjugated Antibodies PN 6607119 - 50 test - 20 μL/test

BUFFER

2.0 mg/mL bovine serum albumin in phosphate-buffered saline containing 0.1% sodium azide.

STATEMENT OF WARNINGS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.

2. Specimens, samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes
4. Do not use antibody beyond the expiration date on the label.
5. Do not expose reagents to strong light during storage or incubation.
6. Use Good Laboratory Practices (GLP) when handling reagent.
7. Harmful if swallowed.
8. After contact with skin, wash immediately with plenty of water.

STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2-8°C. Do not freeze. Minimize exposure to light.

REAGENT PREPARATION

No reconstitution is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18-25°C prior to use.

PROCEDURE

This reagent is designed for flow cytometry. Assay Volume; 20 μL per 5 x 10⁵ cells in one test, or per 100 μL whole blood. A wash maybe required to yield optimal results.

EXAMPLE DATA

The histograms shown are representative of a normal EDTA whole blood sample, stained with CD64-PE/CD16-PC7 multicolor reagent (PN 6607119).

Figures:
Acquisition with a COULTER® EPICS® XL™/XL-MCL™ flow cytometer.

Figure 1:
Side Scatter vs. CD64-PE

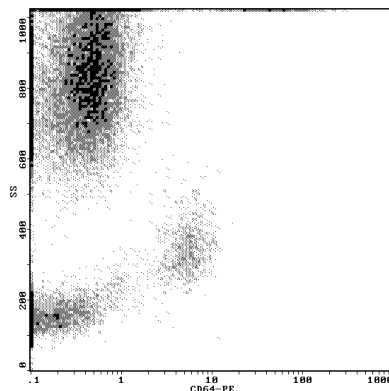


Figure 2:
Side Scatter vs. CD16-PC7

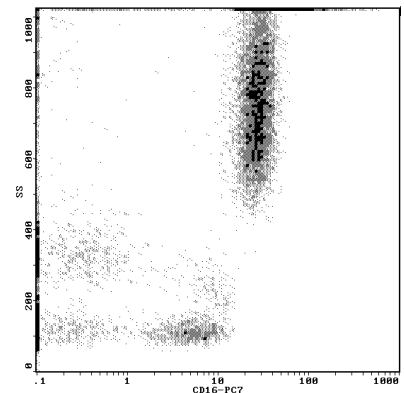
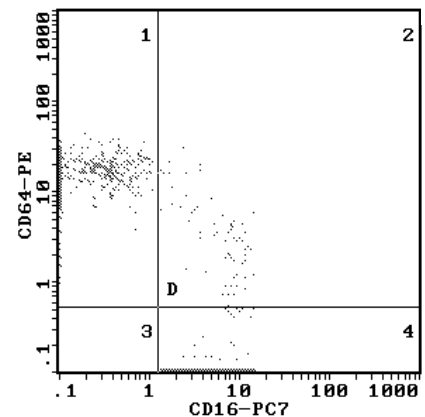


Figure 3:
CD64-PE vs. CD16-PC7 (Monocyte/Lymphocyte Gate)



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