

## 30-1454: Anti-CD16 / FcgammaRIII Monoclonal Antibody (Clone:3G8)

<b>Clonality :</b>	Monoclonal
<b>Clone Name :</b>	3G8
<b>Application :</b>	FACS
<b>Reactivity :</b>	Human
<b>Gene :</b>	FCGR3A
<b>Gene ID :</b>	2214
<b>Uniprot ID :</b>	P08637
<b>Format :</b>	Purified
<b>Alternative Name :</b>	FCGR3A,CD16A,FCG3,FCGR3,IGFR3
<b>Isotype :</b>	Mouse IgG1
<b>Immunogen Information :</b>	Human neutrophils

### Description

CD16 (FcgammaRIII) is a 50-65 kDa glycoprotein serving as a low affinity IgG receptor. Human FcgammaRIII is expressed in two forms - FcgammaRIII-A and -B. FcgammaRIII-A is a transmembrane protein of monocytes, macrophages, NK cells and a subset of T cells. It is associated with FcepsilonRI-gamma subunit and is responsible for antibody-dependent NK cell cytotoxicity. Mast cell FcgammaRIII-A is associated, moreover, with FcepsilonRI-beta subunit. Besides IgG, FcgammaRIII-A can be triggered also by oligomeric IgE. FcgammaRIII-B is a GPI-linked monomeric receptor expressed on neutrophils and is involved in their activation and induction of a proadhesive phenotype.

### Product Info

<b>Amount :</b>	0.1 mg
<b>Purification :</b>	Purified by protein-A affinity chromatography
<b>Storage condition :</b>	Store at 2-8°C. Do not freeze.

### Application Note

**Flow Cytometry** *Recommended dilution:* 6 µg/ml

**Immunoprecipitation Immunohistochemistry (frozen sections)** *Application note:* acetone fixation

**Functional Application** In vitro Stimulation of NK cell proliferation, blocking of IgG binding and phagocytosis, inhibition of cytotoxic activity, in vivo NK cell depletion

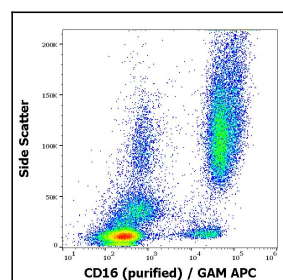


Figure 1: Flow cytometry surface staining pattern of human peripheral whole blood stained using anti-human CD16 (3G8) purified antibody (concentration in sample 2 1/4g/ml, GAM APC).

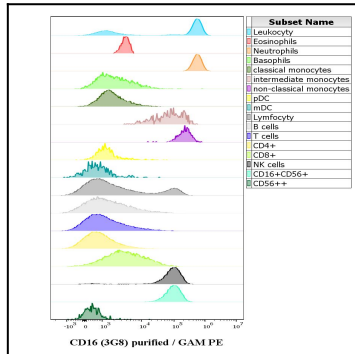


Figure 2: Expression profiling on peripheral blood subsets using anti-human CD16 purified antibody (clone 3G8). HCDM CDMaps standardized procedures (Kuzilkova D et al. Front Immunol. 2022;13:827898) were used for cell isolation and surface staining of blood leukocytes, with the modification of staining protocol using cytometry test tubes. Suspension of blood leukocytes isolated from buffy coats ( $2 \times 10^6$  cells) with residual erythrocytes lysed with  $10\text{Å}$ — diluted EXCELLYSE Live solution (#ED7068) was added to the mixture of anti-human CD16 purified antibody (clone 3G8,  $0.5 \mu\text{g}/\text{ml}$  in stained blood sample) and Monocyte Blocking Buffer (#ED7747), vortexed and incubated for 20 min. Next, samples were centrifuged (670 g, 5 min.), supernatant removed and secondary antibody (GAM PE) was added to sample, vortexed and incubated for 20 min. Next, samples were washed twice (2 ml PBS, 670 g, 5 min.) and then optimized backbone antibody panels (HLDA Innate and HLDA Adaptive) were added to test tubes, vortexed and incubated for 20 min. Next, samples are fixed with 2 ml of  $10\text{Å}$ — diluted EXCELLYSE Easy solution (#ED7066) and incubated for 10 min. Finally, samples were centrifuged (670 g, 5 min.), supernatant removed and the cell pellet was resuspended in  $200 \mu\text{l}$  of PBS for acquisition.

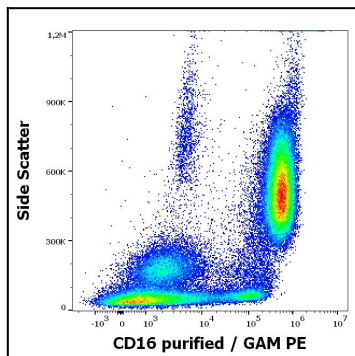


Figure 3: Anti-human CD16 purified antibody (clone 3G8) works in flow cytometry application. Analysis of the antibody staining profile was performed on blood leukocytes isolated from buffy coats. HCDM CDMaps standardized procedures (Kuzilkova D et al. Front Immunol. 2022;13:827898) were used for cell isolation and surface staining of blood leukocytes, with the modification of staining protocol using cytometry test tubes. Mouse monoclonal anti-human CD16 purified antibody (clone 3G8) was used in concentration  $0.5 \mu\text{g}/\text{ml}$  in stained blood sample ( $2 \times 10^6$  cells).

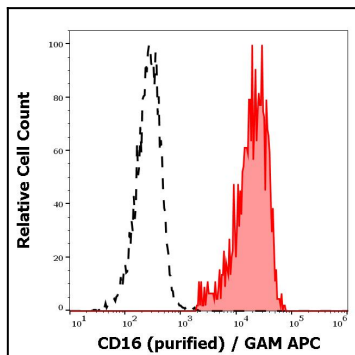


Figure 4: Separation of human CD16 positive lymphocytes (red-filled) from CD16 negative lymphocytes (black-dashed) in flow cytometry analysis (surface staining) of peripheral whole blood stained using anti-human CD16 (3G8) purified antibody (concentration in sample  $2 \frac{1}{4}\text{g}/\text{ml}$ , GAM APC).