# **Technical Data Sheet** Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)

Product Information	
Material Number:	553141
Alternate Name:	Fcy III/II Receptor
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	2.4G2
Immunogen:	Mouse BALB/c Macrophage J774 Cell Line
Isotype:	Rat IgG2b κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing $\leq 0.09\%$ sodium azide.

## Description

The 2.4G2 antibody reacts specifically with a common nonpolymorphic epitope on the extracellular domains of the mouse FcyIII and FcyII receptors. It has also been reported to bind the FcyI receptor (CD64) via its Fc domain. 2.4G2 mAb blocks non-antigen-specific binding of immunoglobulins to the FcyIII and FcyII, and possibly FcyI, receptors in vitro and in vivo. CD16 and/or CD32 are expressed on natural killer cells, monocytes, macrophages, dendritic cells (at low levels), Kupffer cells, granulocytes, mast cells, B lymphocytes, immature thymocytes, and some activated mature T lymphocytes.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.



Two color analysis of the expression of CD16/CD32 on mouse spleen cells and demonstration of FCyR-mediated non-specific staining. Left: BALB/c splenocytes were simultaneously stained with PE-conjugated anti-mouse CD3e mAb 145-2C11 (Cat. No. 553063/553064) and purified 2.4G2 mAb. The staining by 2.4G2 antibody was detected with FITC-conjugated mouse anti-rat Ig, k chain mAb MRK-1 (Cat. No. 553872). Right: BALB/c splenocytes were stained with FITC-conjugated rat anti-mouse CD90.2 (Thy-1.2) mAb 53-2.1 (Cat. No. 553003/553004) in the presence of purified 2.4G2 mAb (filled histogram) and without 2.4G2 mAb (open histogram). Flow cytometry was performed on a BD FACScan™ flow cytometry system

## **Preparation and Storage**

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4° C.

### **Application Notes**

Applicat	ion								
Flow cytometry				Routinely Te	Routinely Tested				
BD Bioscier	nces								
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Blocking	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunoprecipitation	Reported

## **Recommended Assay Procedure:**

To specifically stain cells bearing  $Fc\gamma II$  and  $Fc\gamma III$  receptors for flow cytometric analysis: Incubate cell suspension with this antibody ( $\leq 1 \mu g$ /million cells) followed by an appropriate fluorochrome-conjugated second-step reagent.

To reduce Fc receptor-mediated binding by antibodies of interest or Fc receptor-mediated binding by PE-CY5 tandem dye conjugates to FcyII and FcyIII receptor-bearing mouse cells for flow cytometric analysis:

1. Preincubate cell suspension with Mouse BD Fc Block<sup>TM</sup> purified anti-mouse CD16/CD32 mAb 2.4G2 (eg,  $\leq 1 \mu$ g/million cells in 100  $\mu$ l) at 4° C for 5 minutes.

2. Add antibody of interest directly to preincubated cells in the presence of Mouse BD Fc Block<sup>TM</sup> (ie, Mouse BD Fc Block<sup>TM</sup> need not be washed off before staining cells).

3. If anti-Ig second-step is necessary, a reagent must be chosen which will not bind to Mouse BD Fc  $Block^{TM}$  (eg, rat IgG<sub>2b</sub>,  $\kappa$ ).

For additional information on using Mouse BD Fc Block<sup>TM</sup>, refer to our website protocol at http://www.bdbiosciences.com/pharmingen/protocols/Immunophenotyping.shtml

### **Product Notices**

- 1. 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.
- 2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

#### References

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