Technical Data Sheet

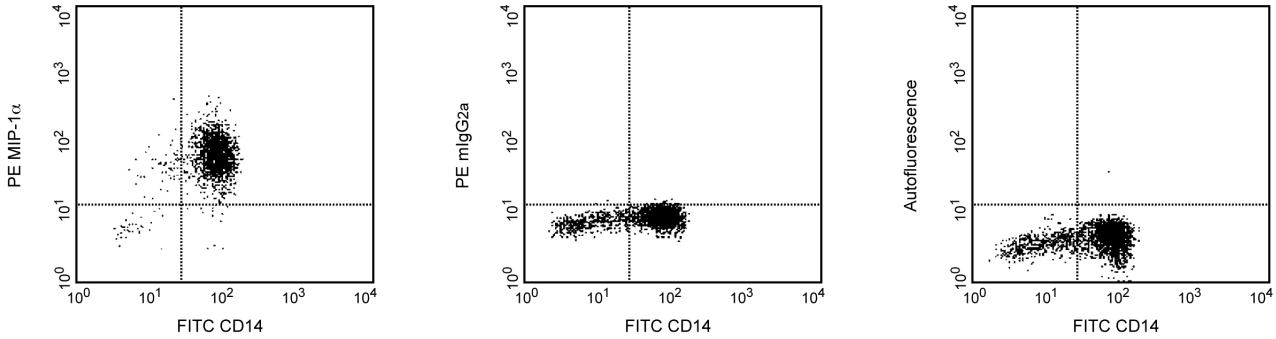
PE Mouse IgG2a, κ Isotype Control

Product Information

Material Number:	**554648**
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	G155-178
Immunogen:	TNP-keyhole limpet hemocyanin
Isotype:	Mouse (BALB/c) IgG2a, κ
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The G155-178 clone has an unknown specificity. Trinitrophenol (TNP), the immunogen, is a hapten not expressed on human, mouse, rat or non-human primate cells. In the absence of specific binding, this antibody may bind non-specifically to immunoglobulin Fc receptors. The immunoglobulin secreted by the G155-178 hybridoma was selected as an mouse IgG2a, κ isotype control following screening for low background binding on a variety of mouse and human tissues.



***Expression of human MIP-1α by stimulated CD14+ human monocytes**. Human PBMC were stimulated for 6 hours with LPS (100 ng/ml final concentration) in the presence of GolgiStop™ (2 µM final concentration; Cat. No. 554724). The PBMC were harvested, stained with FITC-mouse anti-human CD14 monoclonal antibody (FITC-M5E2, Cat. No. 555397), fixed permeabilized, and subsequently stained with either PE-anti-human MIP-1α (Cat. No. 554730, left panel), or PE-mouse IgG2a, κ (Cat. No. 559319; middle panel), by following the Pharmingen staining protocols. The data reflect gating on monocytes, based on forward and scattered light signals. The quadrant markers for the bivariate dot plot were set based on autofluorescence controls (right panel).*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
The antibody was conjugated with R-PE under optimum conditions, and unconjugated antibody and free PE were removed.
Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

Application Notes

Application

Flow cytometry	Routinely Tested
Isotype control	Routinely Tested
Intracellular staining (flow cytometry)	Routinely Tested

Recommended Assay Procedure:

Immunofluorescent Staining and Flow Cytometric Analysis: The FITC- and PE-conjugated G155-178 immunoglobulins (Cat. No. 554647; No. 554648) are suitable mouse IgG2a isotype controls for assessing the level of background staining on paraformaldehyde fixed/saponin-permeabilized rat or human cells for flow cytometric analysis. Use at comparable concentrations to antibody of interest (middle panel). For specific methodology, please visit our website, www.bdbiosciences.com and go to the protocols section or the chapter on intracellular staining in the Immune Function Handbook. The intracellular cytokine staining technique and use of blocking controls are described in detail by C. Prussin and D. Metcalfe.

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Product Notices

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

References

Prussin C, Metcalfe DD. Detection of intracytoplasmic cytokine using flow cytometry and directly conjugated anti-cytokine antibodies. *J Immunol Methods*. 1995; 188(1):117-128. (Methodology: Blocking, Flow cytometry)