

Technical Data Sheet

Alexa Fluor® 488 Mouse IgG1 κ Isotype Control

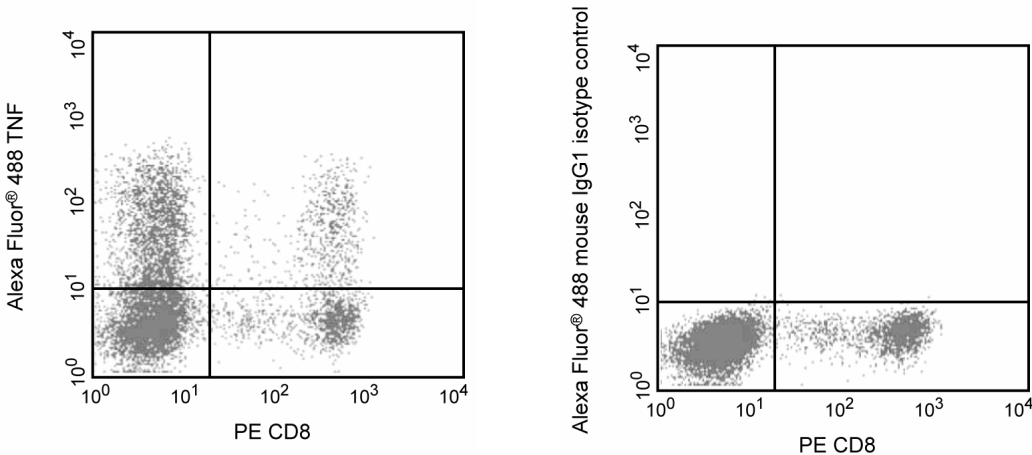
Product Information

Material Number:	557721
Size:	100 tests
Vol. per Test:	5 µl
Clone:	MOPC-21
Isotype:	Mouse IgG1, κ
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The MOPC-21 immunoglobulin is a mouse myeloma protein. The MOPC-21 immunoglobulin was selected as an isotype control following screening for low background on a variety of mouse and human tissues.

Cat. no. 557721 has been optimized for assessing the level of background staining on paraformaldehyde fixed/saponin-permeabilized mouse or human cells for flow cytometric analysis. 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.



**Expression of TNF by stimulated human peripheral blood lymphocytes.** Human peripheral blood mononuclear cells were stimulated for 4 hours with PMA (5 ng/ml, Sigma, Cat. No. P-8139) and Ionomycin (500 ng, Sigma, P-8139) in the presence of Brefeldin A (GolgiPlug, Cat. No. 555029). Cells were harvested, fixed, permeabilized and stained with PE-conjugated mouse anti-human CD8 (PE-RPA-T8, Cat. No. 555367) and either mouse anti-human TNF antibody (Alexa Fluor® 488-MAb11, Cat. No. 557722), (left panel) or Alexa Fluor® 488-MOPC-21 immunoglobulin (Cat. No. 557721) (right panel) by using the staining protocol found in the protocols section at [www.bdbioscience.com](http://www.bdbioscience.com). To demonstrate specificity of staining the binding of Alexa Fluor® 488-MAb11 was blocked by the preincubation of the conjugated antibody with molar excess of recombinant human TNF (Cat. No. 554618) and by preincubation of the fixed permeabilized cells with an excess of unlabelled MAb11 antibody (5 µg, Cat. No. 554510) prior to staining. Dot plots were derived from gated events with the forward and side light scatter characteristics of lymphocytes. The quadrant markers for the bivariate dot plots were set based on the autofluorescence and isotype controls.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. The antibody was conjugated to Alexa Fluor® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was 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)	Tested During Development

Recommended Assay Procedure:

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BD Biosciences

[bdbiosciences.com](http://bdbiosciences.com)  
United States 877.232.8995 Canada 888.259.0187 Europe 32.53.720.550 Japan 0120.8555.90 Asia Pacific 65.6861.0633 Latin America/Caribbean 55.11.5185.9995  
For country-specific contact information, visit [bdbiosciences.com/how\\_to\\_order/](http://bdbiosciences.com/how_to_order/)  
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**Immunofluorescent Staining and Flow Cytometric Analysis:** The FITC-, PE-, APC-, PE-Cy7-, Alexa Fluor® 488-, and Alexa Fluor® 647-MOPC-21 immunoglobulins (Cat. No. 554679, 559320, 554681, 557646, 557721, and 557732) are suitable mouse IgG1κ isotype controls for assessing the level of background staining on paraformaldehyde fixed/saponin-permeabilized mouse or human cells for flow cytometric analysis.

### Suggested Companion Products

Catalog Number	Name	Size	Clone
554715	BD Cytofix/Cytoperm Plus Kit (with BD GolgiStop)	250 tests	(none)
555028	BD Cytofix/Cytoperm Plus Kit (with BD GolgiPlug)	250 tests	(none)
555029	Protein Transport Inhibitor (Containing Brefeldin A)	1.0 ml	(none)

### Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  cells in a 100-μl experimental sample (a test).
2. Please refer to [www.bdbiosciences.com/pharmingen/protocols](http://www.bdbiosciences.com/pharmingen/protocols) for technical protocols.
3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
4. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
5. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
6. 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.
7. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
8. Alexa Fluor is a registered trademark of Molecular Probes, Inc., Eugene, OR.

### 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: Flow cytometry)