

Technical Data Sheet

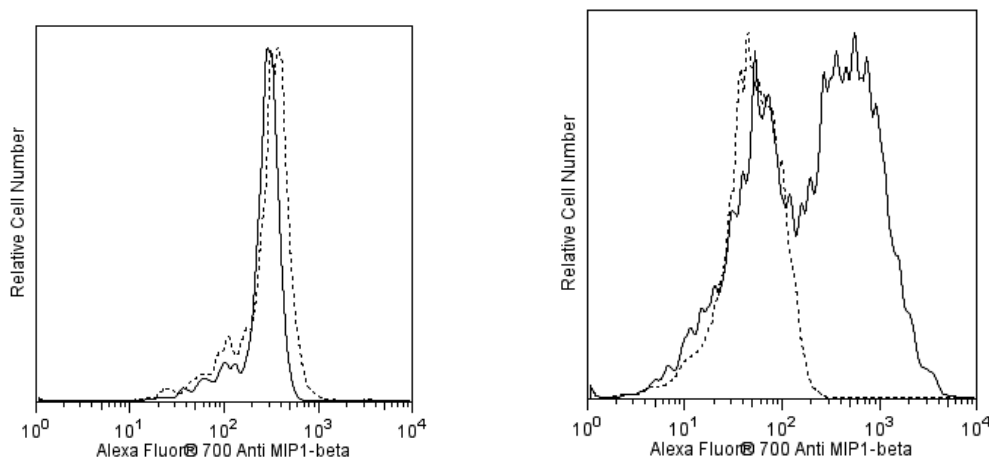
Alexa Fluor® 700 Mouse Anti-Human MIP-1β

Product Information

Material Number:	561278
Alternate Name:	Macrophage inflammatory protein 1-beta; CCL4; C-C motif chemokine 4; LAG-1
Size:	50 tests
Vol. per Test:	5 µl
Clone:	D21-1351
Immunogen:	Recombinant Human MIP-1β
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Storage Buffer:	Aqueous buffered solution containing protein stabilizer and ≤0.09% sodium azide.

Description

The D21-1351 monoclonal antibody specifically binds to the human CC chemokine, MIP-1β (macrophage inflammatory protein-1β). Human MIP-1β shares approximately 75% homology with mouse MIP-1β at the amino acid level. Expression of MIP-1β in human peripheral blood cells is induced by proinflammatory and mitogenic stimuli. MIP-1β is a chemoattractant for monocytes and lymphocytes. Human MIP-1β binds to receptors, CCR5 and CCR8. The human MIP-1β gene has been mapped to chromosome 17q11. The immunogen used to generate D21-1351 hybridoma was recombinant human MIP-1β.



Flow cytometric analysis of MIP-1β expressed in human peripheral blood mononuclear cells (PBMC). Human PBMC were either unstimulated (Left Panel) or stimulated (Right Panel) with 20 ng/mL Recombinant Human IFN-γ (Cat. No. 554616) for one hour followed by overnight incubation with 1 µg/mL LPS (Sigma-Aldrich, Cat. No. L-8272) in the presence of 2 µM BD GolgiStop™ Protein Transport Inhibitor (Cat. No. 554724). The PBMC were harvested, fixed with BD Cytotfix™ Fixation Buffer (Cat. No. 554655) and permeabilized with BD Perm/Wash™ Buffer (Cat. No. 554723). The cells were then stained with either Alexa Fluor® 700 Mouse IgG1, κ Isotype Control (Cat. No. 557882; dashed line histogram) or with the Alexa Fluor® 700 Mouse Anti-Human MIP-1β antibody (Cat. No. 561278; solid line histogram). The fluorescence histograms were derived from gated events with the forward and side light-scatter characteristics of viable monocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

The antibody was conjugated to Alexa Fluor® 700 under optimum conditions, and unreacted Alexa Fluor® 700 was removed.

Store undiluted at 4°C and protected from prolonged exposure to light. Do not freeze.

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Application Notes

Application

Intracellular staining (flow cytometry)

Routinely Tested

Suggested Companion Products

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
554616	Recombinant Human IFN- γ	25 μ g	(none)
554724	Protein Transport Inhibitor (Containing Monensin)	0.7 ml	(none)
554655	Fixation Buffer	100 ml	(none)
554723	Perm/Wash Buffer	100 ml	(none)
557882	Alexa Fluor® 700 Mouse IgG1, κ Isotype Control	0.1 mg	MOPC-21
554656	Stain Buffer (FBS)	500 ml	(none)

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100- μ l experimental sample (a test).
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
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® 700 has an adsorption maximum of ~700nm and a peak fluorescence emission of ~720nm. Before staining cells with this reagent, please confirm that your flow cytometer is capable of exciting the fluorochrome and discriminating the resulting fluorescence.
6. Alexa Fluor® is a registered trademark of Molecular Probes, Inc., Eugene, OR.
7. 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.
8. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.

References

Bernardini G, Hedrick J, Sozzani S. Identification of the CC chemokines TARC and macrophage inflammatory protein-1 beta as novel functional ligands for the CCR8 receptor. *J Immunol.* 1998; 28(2):582-588. (Biology)

Combadiere C, Ahuja SK, Tiffany HL, Murphy PM. Cloning and functional expression of CC CKR5, a human monocyte CC chemokine receptor selective for MIP-1(alpha), MIP-1(beta), and RANTES. *J Leukoc Biol.* 1996; 60(1):147-152. (Biology)

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, IC/FCM Block)

Raport CJ, Gosling J, Schweickart VL, Gray PW, Charo IF. Molecular cloning and functional characterization of a novel human CC chemokine receptor (CCR5) for RANTES, MIP-1beta, and MIP-1alpha. *J Biol Chem.* 1996; 271(29):17161-17166. (Biology)