

Technical Data Sheet

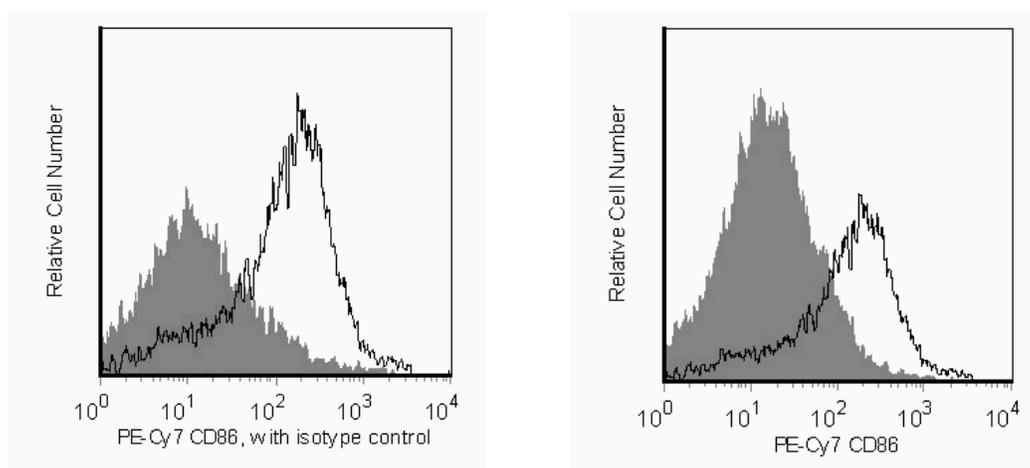
PE-Cy™7 Rat Anti-Mouse CD86

Product Information

Material Number:	560582
Alternate Name:	B7-2
Size:	50 µg
Concentration:	0.2 mg/ml
Clone:	GL1
Immunogen:	Mouse (CBA/Ca) LPS-activated splenic B Cells
Isotype:	Rat (LOU) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The GL1 antibody has been reported to react with the B7-2 (CD86) costimulatory molecule expressed on a broad spectrum of leukocytes, including B lymphocytes, T lymphocytes, thioglycollate-induced peritoneal macrophages, dendritic cells and astrocytes. CD86 is expressed at low levels by freshly explanted peripheral B and T cells, and its expression is substantially increased by a variety of T cell- and B cell-specific stimuli with a peak expression after 18-42 hours of culture. In contrast to most naive CD4+ T cells, memory CD4+ T cells express B7-2, both at the mRNA and protein level. CD86, a ligand for CD28 and CD152 (CTLA-4), is one of the accessory molecules that plays an important role in T cell-B cell costimulatory interactions. It has been shown to be involved in immunoglobulin class-switching and triggering of mouse NK cell-mediated cytotoxicity. CD80 (B7-1) is an alternate ligand for CD28 and CD152 (CTLA-4). GL1 antibody reportedly blocks MLR and stimulation of T cells by natural antigen-presenting cells. In addition, a mixture of anti-B7-1 and anti B7-2 (GL1) mAbs reportedly inhibits the in vitro interaction of CTLA-4 with its ligand and the in vivo priming of cytotoxic T lymphocytes.



Flow cytometric analysis of CD86 on mouse splenocytes. **Left Panel:** LPS-stimulated C57BL/6 splenocytes (72 hr) were stained either with a PE-Cy™7 Rat IgG2a, κ isotype control (shaded) or with the PE-Cy™7 Rat Anti-Mouse CD86 antibody (unshaded). **Right Panel:** Freshly isolated unstimulated C57BL/6 splenocytes (shaded) and LPS-stimulated C57BL/6 splenocytes (72 hr) (unshaded) were stained with the PE-Cy™7 Rat Anti-Mouse CD86 antibody. Histograms were derived from gated events based on light scattering characteristics for lymphocytes. Flow cytometry was performed on a BD™ LSR II flow cytometry system.

Preparation and Storage

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

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

The antibody was conjugated with PE-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
552784	PE-Cy TM 7 Rat IgG2a, κ Isotype Control	0.1 mg	R35-95
553141	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block TM)	0.1 mg	2.4G2

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. An isotype control should be used at the same concentration as the antibody of interest.
3. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BDTM Stabilizing Fixative (Cat. No. 338036).
4. Cy is a trademark of Amersham Biosciences Limited. This conjugated product is sold under license to the following patents: US Patent Nos. 5,486,616; 5,569,587; 5,569,766; 5,627,027.
5. This product is subject to proprietary rights of Amersham Biosciences Corp. and Carnegie Mellon University and made and sold under license from Amersham Biosciences Corp. This product is licensed for sale only for research. It is not licensed for any other use. If you require a commercial license to use this product and do not have one return this material, unopened to BD Biosciences, 10975 Torreyana Rd, San Diego, CA 92121 and any money paid for the material will be refunded.
6. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
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. Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem fluorochrome conjugate; therefore, we recommend that special precautions be taken (such as wrapping vials, tubes, or racks in aluminum foil) to prevent exposure of conjugated reagents, including cells stained with those reagents, to room illumination.
9. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
10. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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