Technical Data Sheet

PE-Cy[™]5 Rat IgG2a, κ Isotype Control

Product Information

Material Number: 551066 Size: 100 tests 20 µl Vol. per Test: R35-95 Clone:

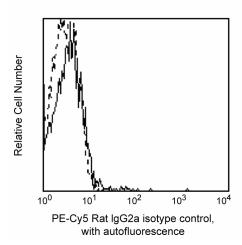
Mouse Pooled Immunoglobulin Immunogen:

Isotype: Rat (LOU) IgG2a, ĸ

Storage Buffer: Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The R35-95 hybridoma was generated by hybridization of Y3 myeloma cells with spleen cells from LOU rats immunized with mouse immunoglobulins. The R35-95 hybridoma produces rat IgG2a, κ immunoglobulin that has no measurable reactivity with mouse immunoglobulins. The R35-95 immunoglobulin was selected as an isotype control following screening for low background binding on a variety of mouse and human tissues.



Profile of peripheral blood lymphocytes analyzed by flow cytometry

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-Cy5 (formerly known as BD Cy-ChromeTM) under optimum conditions, and unconjugated antibody and free PE-Cy5 were removed.

Application Notes

Application

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	Flow cytometry	Routinely Tested
	Isotype control	Routinely Tested

Product Notices

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10⁶ cells in a 100-μl experimental sample (a test).
- 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.
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- PE-Cy5 is optimized for use with a single argon ion laser emitting 488-nm light. Because of the broad absorption spectrum of the PE-Cy5 tandem fluorochrome, extra care must be taken when using dual-laser cytometers which may directly excite both PE and Cy5TM.

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- 5. PE-Cy5 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by the 488 nm light of an Argon ion laser and serves as an energy donor, coupled to the cyanine dye Cy5, which acts as an energy acceptor and fluoresces at 670 nm. BD Biosciences Pharmingen has maximized the fluorochrome energy transfer in PE-Cy5, thus maximizing its fluorescence emission intensity, minimizing residual emission from PE, and minimizing lot-to-lot variation.
- 6. PE-Cy5 tandem fluorochromes have been reported to bind some classes of human macrophages and granulocytes via Fc receptors, and PE has been reported to bind to mouse B lymphocytes via Fc receptors. Preincubation of mouse leukocytes with Mouse BD Fc Block™ purified anti-mouse CD16/CD32 mAb 2.4G2 can reduce the non-specific binding of PE-Cy5-conjugated reagents to mouse B cells. However, PE-Cy5 conjugated reagents should not be used to stain splenocytes of SJL, NOD, and MRL mice as B lymphocytes and/or other leukocytes have been reported to non-specifically stain regardless of the use of Mouse BD Fc Block™ (the CD72c complex has been implicated for PE-Cy5 binding in these strains). Reagents conjugated to PE, PerCP, PerCP-Cy5.5, APC, and APC-Cy7 tandem fluorochrome can be used on leukocytes from these mouse strains.
- 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. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 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.

References

van Vugt MJ, van den Herik-Oudijk IE, van de Winkle JG. Binding of PE-CY5 conjugates to the human high-affinity receptor for IgG (CD64). *Blood.* 1996; 88(6):2358-2361. (Biology)

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