## Technical Data Sheet

# PE-Cy™5 Rat Anti-Mouse CD45

#### **Product Information**

**Material Number:** 561870

Alternate Name: Leukocyte Common Antigen (LCA), Ly-5, T200

Size 0.2 mg/ml Concentration: Clone: 30-F11

Immunogen: Mouse Thymus / Spleen Isotype: Rat (LOU) IgG2b, K Reactivity: QC Testing: Mouse

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

#### Description

The 30-F11 clone has been reported to react with all isoforms and both alloantigens of CD45, which is found on hematopoietic stem cells and all cells of hematopoietic origin, except erythrocytes. CD45 is a transmembrane glycoprotein which is expressed at high levels on the cell-surface, and its presence distinguishes leukocytes from non-hematopoietic cells. CD45 is a member of the Protein Tyrosine Phosphatase (PTP) family, where the intracellular carboxy-terminal region contains two PTP catalytic domains, and the extracellular region is highly variable due to alternative splicing of exons 4, 5, and 6 (designated as A, B, and C, respectively). CD45 isoforms play complex roles in T-cell and B-cell antigen receptor signal transduction and the CD45 isoforms detected in the mouse are cell type-, maturation-, and activation state-specific.

### **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-Chrome<sup>TM</sup>) under optimum conditions, and unconjugated antibody and free PE-Cy5 were removed.

#### **Application Notes**

#### Application

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#### **Recommended Assay Procedure:**

Flow Cytometry: 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. Refer to our website

http://www.bdbiosciences.com/support/resources/index.jsp for more information on mouse and rat flow sample preparation and the uses of Mouse BD Fc BlockTM.

### **Suggested Companion Products**

Catalog Number	Name	Size	Clone	
553990	PE-Cy <sup>TM</sup> 5 Rat IgG2b, κ Isotype Control	0.1 mg	A95-1	
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2	
554656	Stain Buffer (FBS)	500 ml	(none)	

#### **Product Notices**

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
- For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
- 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.
- 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.
- 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.
- 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.

## **BD Biosciences**

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- 8. 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<sup>TM</sup> 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<sup>TM</sup> (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.
- 9. 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.
- 10. 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.

#### References

Johnson P, Maiti A. CD45: A family of leukocyte-specific cell surface glycoproteins. In: Herzenberg LA, Weir DM, Blackwell C, ed. Weir's Handbook of Experimental Immunology, Vol 2. Cambridge: Blackwell Science; 1997:62.1-62.16. (Biology)

Lagasse E, Connors H, Al-Dhalimy M, et al. Purified hematopoietic stem cells can differentiate into hepatocytes in vivo. *Nat Med.* 2000; 6(11):1212-1213. (Biology) Ledbetter JA, Herzenberg LA. Xenogeneic monoclonal antibodies to mouse lymphoid differentiation antigens. *Immunol Rev.* 1979; 47:63-90. (Immunogen) Takizawa F, Kinet JP, Adamczewski M. Binding of phycoerythrin and its conjugates to murine low affinity receptors for immunoglobulin G. *J Immunol Methods*. 1993; 162(2):269-272. (Methodology)

Thomas ML. The leukocyte common antigen family. Annu Rev Immunol. 1989; 7:339-369. (Biology)

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)

Waggoner AS, Ernst LA, Chen CH, Rechtenwald DJ. PE-CY5. A new fluorescent antibody label for three-color flow cytometry with a single laser. *Ann N Y Acad Sci.* 1993; 677:185-193. (Biology)

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