## **Technical Data Sheet**

# PE-Cy<sup>™</sup>5 Mouse Anti-Human CD25

#### **Product Information**

Material Number: 555433

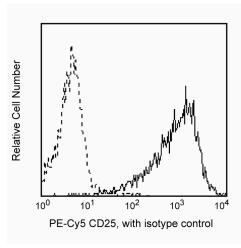
Alternate Name: IL-2R; IL-2Rα; TCGFR; TAC antigen; p55

Workshop: IV A053

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

#### Description

The M-A251 monoclonal antibody specifically binds to the 55 kDa type I transmembrane glycoprotein known as the low-affinity interleukin-2 receptor alpha chain subunit (IL-2R $\alpha$ ). CD25 is expressed on regulatory T cells and on activated lymphocytes (T and B) and monocytes. It associates with the IL-2R $\beta$ /CD122 and the IL-2R $\gamma$ /CD132 receptor chains to form the high-affinity IL-2R complex. CD25 expression on T and B lymphocytes is upregulated by antigenic or mitogenic stimulation. Soluble CD25/IL-2R $\alpha$  is produced as a consequence of lymphocyte stimulation and is found in biological fluids following inflammatory responses.



Profile of PHA-stimulated (3 days) peripheral blood lymphocytes analyzed on a FACScan (BDIS, San Jose, CA)

# **Preparation and Storage**

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.

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

### **Application Notes**

Application Routinely Tested

# **Suggested Companion Products**

 Catalog Number
 Name
 Size
 Clone

 555750
 PE-Cy<sup>TM</sup>5 Mouse IgG1 κ Isotype Control
 100 tests
 MOPC-21

#### **Product Notices**

- This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1 × 10<sup>6</sup> cells in a 100-µl experimental sample (a test).
- 2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- 3. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
- 4. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

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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 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 Cy5<sup>TM</sup>.
- 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.
- 8. 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.
- 9. 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.
- 10. 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.
- 11. Source of all serum proteins is from USDA inspected abattoirs located in the United States.

#### References

Knapp W, Dorken B, Rieber EP, et al, ed. Leucocyte Typing IV. New York: Oxford University Press; 1989:1-1208. (Clone-specific)
Schlossman SF, Boumsell L, Gilks W, et al, ed. Leukocyte Typing V: White Cell Differentiation Antigens. New York: Oxford University Press; 1995. (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)

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