Technical Data Sheet

PE-Cy™5 Mouse Anti-Human CD45RA

Product Information

Material Number: 552888 Size: 0.1 mg0.2 mg/ml**Concentration:** 5H9 Clone: Isotype: Mouse IgG1, κ

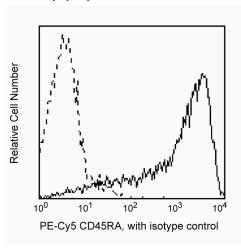
Reactivity: Human

QC Testing: Baboon or Cynomolgus or Rhesus

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

Description

The 5H9 monoclonal antibody specifically binds to the human form of the leukocyte common antigen (LCA), CD45RA. This clone also crossreacts with CD45RA expressed by peripheral blood lymphocytes, monocytes and some granulocytes from rhesus macaque, baboon, and cynomolgus monkeys. The observed frequency of CD45RA positive lymphocytes is higher in the non-human primate compared to what is seen with normal human donor lymphocytes.



Profile of anti-CD45RA (5H9) reactivity on peripheral blood lymphocytes of rhesus macaque (Macaca mulatta) monkey 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

Flow cytometry Routinely Tested

- Since applications vary, each investigator should titrate the reagent to obtain optimal results.
- An isotype control should be used at the same concentration as the antibody of interest.
- 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.
- 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.
- 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.

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- 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.
- 7. 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.
- 8. 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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- 11. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
- 12. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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Reimann KA, Waite BC, Lee-Parritz DE, et al. Use of human leukocyte-specific monoclonal antibodies for clinically immunophenotyping lymphocytes of rhesus monkeys. *Cytometry*. 1994; 17(1):102-108. (Biology)

Sopper S, Stahl-Hennig C, Demuth M, Johnston IC, Dorries R, ter Meulen V. Lymphocyte subsets and expression of differentiation markers in blood and lymphoid organs of rhesus monkeys. *Cytometry*. 1997; 29(4):351-362. (Biology)

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