

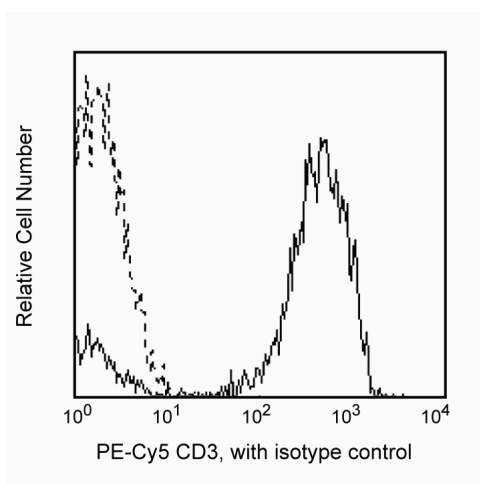
Technical Data Sheet

PE-Cy™5 Mouse Anti-Human CD3**Product Information**

Material Number:	555341
Size:	100 tests
Vol. per Test:	20 µl
Clone:	HIT3a
Isotype:	Mouse IgG2a, κ
Reactivity:	QC Testing: Human
Workshop:	V 5T-CD03.05
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

Reacts with the human ε-chain, a 20 kDa subunit of CD3/T cell antigen receptor complex found on 70-80% of normal human peripheral blood lymphocytes and 60-85% of thymocytes. Studies from the HLDA Workshop show this antibody to be mitogenic when used in conjunction with pokeweed mitogen. CD3 plays a role in signal transduction during antigen recognition. HIT3a antibody does not stain intracellular CD3 unlike the other CD3 clone, UCHT1.



Profile of peripheral blood lymphocytes analyzed on a FACSscan (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™) 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**

Flow cytometry	Routinely Tested
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Suggested Companion Products

Catalog Number	Name	Size	Clone
555575	PE-Cy™5 Mouse IgG2a, κ Isotype Control	100 tests	G155-178

Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use 1×10^6 cells in a 100-µl experimental sample (a test).
2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
3. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
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.

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5. 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™.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
7. 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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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

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