Technical Data Sheet

PE-CF594 Mouse Anti-Human CD3

Product Information

Material Number: 562280

Alternate Name: CD3e; CD3E; T3E; TCRE; T-cell surface antigen T3/Leu-4 epsilon

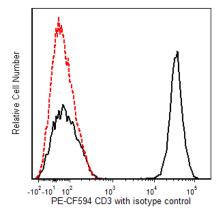
Workshop: III 471

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

Description

The UCHT1 monoclonal antibody specifically binds to the human CD3ε-chain, a 20-kDa subunit of the CD3/T cell antigen receptor complex. CD3ε is expressed on 70-80% of normal human peripheral blood lymphocytes and 60-85% of thymocytes. Studies from the HLDA Workshop show that this antibody is mitogenic for CD3ε-positive cells when used in conjunction with costimulatory agents such as pokeweed mitogen or anti-CD28 antibody. CD3 plays a central role in signal transduction during antigen recognition. The UCHT1 antibody stains both surface and intracellular CD3ε unlike the other CD3 clone, HIT3a, that stains only extracellular CD3ε.

This antibody is conjugated to BD HorizonTM PE-CF594, which has been developed exclusively by BD Biosciences as a better alternative to PE-Texas Red. PE-CF594 excites and emits at similar wavelengths to PE-Texas Red yet exhibits improved brightness and spectral characteristics. Due to PE having maximal absorption peaks at 496 nm and 564 nm, PE-CF594 can be excited by the blue (488-nm), green (532-nm) and yellow-green (561-nm) lasers and can be detected with the same filter set as PE-Texas Red (eg 610/20-nm filter).



Flow cytometric analysis of CD3 expression on human peripheral blood lymphocytes. Human whole blood was stained with the BD Horizon™ PE-CF594 Mouse Anti-Human CD3 antibody (Cat. No. 562280/562310; solid line histogram) or with a BD Horizon™ PE-CF594 Mouse IgG1, κ Isotype Control (Cat. No. 562292; dashed line histogram). The erythrocytes were lysed with BD Pharm Lyse™ Lysing Buffer (Cat. No. 555899). The fluorescence histograms were derived from events with the forward and side light-scatter characteristics of viable lymphocytes. Flow cytometry was performed using a BD™ LSR II Flow Cytometer System.

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 BD Horizon™ PE-CF594 under optimum conditions, and unconjugated antibody and free PE-CF594 were removed.

Application Notes

Application

Flow cytometry Routinely Tested

Suggested Companion Products

Catalog Number	Name	Size	Clone	
562292	PE-CF594 Mouse IgG1, κ Isotype Control	0.1 mg	X40	
554656	Stain Buffer (FBS)	500 ml	(none)	
555899	Lysing Buffer	100 ml	(none)	

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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).
- 2. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
- Because of the broad absorption spectrum of the tandem fluorochrome, extra care must be taken when using multi-laser cytometers, which
 may directly excite both PE and CFTM594.
- 4. An isotype control should be used at the same concentration as the antibody of interest.
- 5. 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.
- 6. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
- CFTM is a trademark of Biotium, Inc.
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- 9. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 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. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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Knapp W, Dorken B, Rieber EP, et al, ed. Leucocyte Typing IV. New York: Oxford University Press; 1989:1-1208. (Clone-specific)

Lanier LL, Allison JP, Phillips JH. Correlation of cell surface antigen expression on human thymocytes by multi-color flow cytometric analysis: implications for differentiation. *J Immunol.* 1986; 137(8):2501-2507. (Biology)

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