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Technical Data Sheet

PE-CF594 Mouse Anti-Human CD8

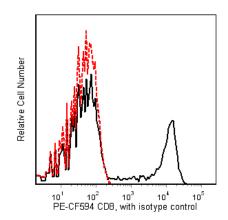
Product Information

Material Number:	562282	
Alternate Name:	CD8a; CD8A; CD8 alpha; Leu2; MAL; T8; p32	
Size:	100 Tests	
Vol. per Test:	5 μl	
Clone:	RPA-T8	
Isotype:	Mouse IgG1, ĸ	
Reactivity:	QC Testing: Human	
	Tested in Development: Rhesus, Cynomolgus, Baboon	
Workshop:	IV T171; V T-CD08.03; VI 6T-CD8.1, 6T-081	
Storage Buffer:	Aqueous buffered solution containing BSA and $\leq 0.09\%$ sodium a	

Description

The RPA-T8 monoclonal antibody specifically binds to CD8 alpha (CD8 α). CD8 α is a type I transmembrane glycoprotein and a member of the immunoglobulin superfamily. CD8 α is expressed by the majority of thymocytes, by subpopulations of $\alpha\beta$ T cells and $\gamma\delta$ T cells and by some NK cells. Cell surface CD8 α is expressed either as a disulfide-linked homodimer (CD8 $\alpha\alpha$) or as a heterodimer (CD8 $\alpha\beta$) when disulfide-bonded to a CD8 beta chain (CD8 β). CD8-positive $\alpha\beta$ T cells coexpress both CD8 $\alpha\alpha$ homodimers and CD8 $\alpha\beta$ heterodimers whereas some $\gamma\delta$ T cells and NK cells express CD8 $\alpha\alpha$ homodimers. CD8 plays important roles in T cell activation and selection. The extracellular IgSF domain of CD8 α binds to a non-polymorphic determinant on HLA class I molecules ($\alpha3$ domain) and enables CD8 to function as a co-receptor with MHC class I-restricted TCR during T cell recognition of antigen. The cytoplasmic domain of CD8 α associates with Lck, a Src family protein tyrosine kinase that is involved in intracellular signaling. The RPA-T8 and HIT8a monoclonal antibodies are not cross-blocking. This clone has been reported to react with a subset of peripheral blood lymphocytes, but not monocytes nor granuloyctes, of baboon and both rhesus and cynomolgus macaque monkey. In general, a higher frequency of CD8+ and CD4+CD8+ lymphocytes are observed in non-human primates compared to normal human donors.

This antibody is conjugated to BD Horizon 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 CD8 expression on human peripheral blood lymphocytes. Human whole blood was stained with the BD Horizon™ PE-CF594 Mouse Anti-Human CD8 antibody (Cat. No. 562282/562311; 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.

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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)

Product Notices

- 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 1. sample (a test).
- 2. An isotype control should be used at the same concentration as the antibody of interest.
- 3. 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.
- 4. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
- 5. When excited by the yellow-green (561-nm) laser, the fluorescence may be brighter than when excited by the blue (488-nm) laser.
- 6. 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 CF™594.
- Please observe the following precautions: Absorption of visible light can significantly alter the energy transfer occurring in any tandem 7. 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.
- 8. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
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- 11. Texas Red is a registered trademark of Molecular Probes, Inc., Eugene, OR.
- For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at 12. www.bdbiosciences.com/colors
- 13. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Knapp W, Dörken B, Gilks WR, et al, ed. Leucocyte Typing IV. New York, NY: Oxford University Press; 1989:1-1182. (Biology) Schlossman SF, Boumsell L, Gilks W, et al, ed. Leucocyte Typing V. New York: Oxford University Press; 1995. (Clone-specific)

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