Technical Data Sheet

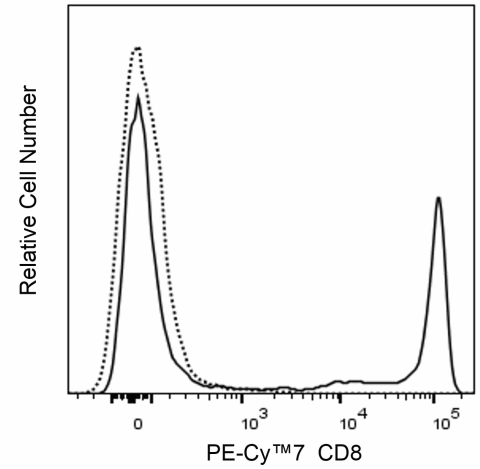
PE-Cy™7 Mouse Anti-Human CD8

Product Information

Material Number:	**566859**
Alternate Name:	CD8α; CD8A; CD8 alpha; Leu2a; MAL; T8; p32
Size:	25 Tests
Vol. per Test:	5 µl
Clone:	HIT8a
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human
Workshop:	V 5T-CD08.10
Storage Buffer:	Aqueous buffered solution containing BSA and ≤0.09% sodium azide.

Description

The HIT8a monoclonal antibody specifically binds to CD8a (CD8α). CD8α is a type I transmembrane glycoprotein and a member of the immunoglobulin superfamily. CD8α is expressed by the majority of thymocytes, by subpopulations of αβ T cells and γδ T cells and by some NK cells. Cell surface CD8α is expressed either as a disulfide-linked homodimer (CD8αα) or as a heterodimer (CD8αβ) when disulfide-bonded to a CD8 beta chain (CD8β). CD8-positive αβ T cells coexpress both CD8αα homodimers and CD8αβ heterodimers whereas some γδ T cells and NK cells express CD8αα homodimers. CD8 plays important roles in T cell activation and selection. The extracellular IgSF domain of CD8α binds to a non-polymorphic determinant on HLA class I molecules (α3 domain) and enables CD8 to function as a coreceptor with MHC class I-restricted TCR during T cell recognition of antigen. The cytoplasmic domain of CD8α associates with Lck, a Src family protein tyrosine kinase that is involved in intracellular signaling. Clones HIT8a and RPA-T8 are not cross-blocking.



Flow cytometric analysis of CD8 expression on human peripheral blood lymphocytes. *Whole blood was stained with either PE-Cy™7 Mouse IgG1 κ Isotype Control (Cat. No. 565573; dashed line histogram) or PE-Cy™7 Mouse Anti-Human CD8 antibody (Cat. No. 566858/566859; solid line histogram). The erythrocytes were lysed with BD FACS™ Lysing Solution (Cat. No. 349202). The histogram showing CD8 expression [or Ig Isotype control staining] was derived from gated events with the forward and side-light scatter characteristics of intact cells. Flow cytometry and data analysis were performed using a BD LSRFortessa™ X-20 Cell Analyzer System and FlowJo™ software.*

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-Cy7 under optimum conditions, and unconjugated antibody and free PE-Cy7 were removed.

Application Notes

Application

Flow cytometry	Routinely Tested

BD Biosciences

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Suggested Companion Products

Catalog Number	Name	Size	Clone
566858	PE-Cy™7 Mouse Anti-Human CD8	100 Tests	HIT8a
565573	PE-Cy™7 Mouse IgG1 κ Isotype Control	50 µg	MOPC-21
555899	Lysing Buffer	100 mL	(none)
349202	BD FACS™ Lysing Solution	100 mL	(none)
554656	Stain Buffer (FBS)	500 mL	(none)
554657	Stain Buffer (BSA)	500 mL	(none)

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. 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. 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 Multicolor Flow Cytometry web page at www.bdbiosciences.com/colors.
7. Warning: Some APC-Cy7 and PE-Cy7 conjugates show changes in their emission spectrum with prolonged exposure to formaldehyde. If you are unable to analyze fixed samples within four hours, we recommend that you use BD™ Stabilizing Fixative (Cat. No. 338036).
8. PE-Cy7 is a tandem fluorochrome composed of R-phycoerythrin (PE), which is excited by 488-nm light and serves as an energy donor, coupled to the cyanine dye Cy7, which acts as an energy acceptor and fluoresces maximally at 780 nm. PE-Cy7 tandem fluorochrome emission is collected in a detector for fluorescence wavelengths of 750 nm and higher. Although every effort is made to minimize the lot-to-lot variation in the efficiency of the fluorochrome energy transfer, differences in the residual emission from PE may be observed. Therefore, we recommend that individual compensation controls be performed for every PE-Cy7 conjugate. PE-Cy7 is optimized for use with a single argon ion laser emitting 488-nm light, and there is no significant overlap between PE-Cy7 and FITC emission spectra. When using dual-laser cytometers, which may directly excite both PE and Cy7, we recommend the use of cross-beam compensation during data acquisition or software compensation during data analysis.
9. Cy is a trademark of GE Healthcare.
10. Please refer to http://regdocs.bd.com to access safety data sheets (SDS).
11. Please refer to www.bdbiosciences.com/us/s/resources for technical protocols.

References

Han L, Chen J, Ding K, et al. Efficient generation of bispecific IgG antibodies by split intein mediated protein trans -splicing system. *Sci Rep* . 2017; 7(1):8360. (Clone-specific: Flow cytometry)

Schlossman SF. Stuart F. Schlossman .. et al., ed. *Leucocyte typing V : white cell differentiation antigens : proceedings of the fifth international workshop and conference held in Boston, USA, 3-7 November, 1993.* Oxford: Oxford University Press; 1995(Clone-specific: Flow cytometry)