

## Technical Data Sheet

## V500 Mouse Anti-Human CD8

## Product Information

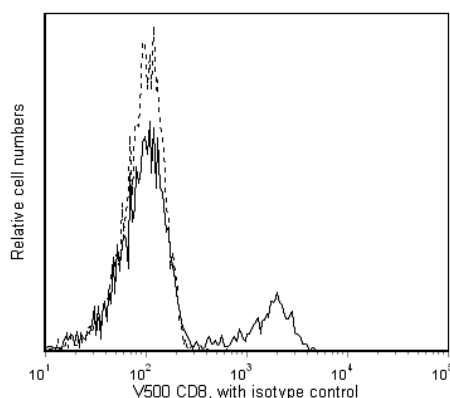
<b>Material Number:</b>	<b>560774</b>
<b>Alternate Name:</b>	CD8α; CD8A; CD8 alpha; Leu2; MAL; T8; p32
<b>Size:</b>	100 Tests
<b>Vol. per Test:</b>	5 µl
<b>Clone:</b>	RPA-T8
<b>Isotype:</b>	Mouse IgG1, κ
<b>Reactivity:</b>	QC Testing: Human Tested in Development: Rhesus, Cynomolgus, Baboon IV T171; V T-CD08.03; VI 6T-CD8.1, 6T-081
<b>Workshop:</b>	
<b>Storage Buffer:</b>	Aqueous buffered solution containing protein stabilizer, glycerol and ≤0.09% sodium azide.

## Description

The RPA-T8 monoclonal antibody specifically binds to CD8 alpha (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 co-receptor 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. 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 granulocytes, 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.

The antibody is conjugated to BD Horizon V500, which has been developed for use in multicolor flow cytometry experiments and is available exclusively from BD Biosciences. It is excited by the Violet laser with an Ex max of 415 nm and Em Max at 500 nm. BD Horizon V500 conjugates emit at a similar wavelength to Amcyan yet exhibit reduced spillover into the FITC channel. For more information on BD Horizon V500, visit [bdbiosciences.com/colors](http://bdbiosciences.com/colors).

When compensating dyes in this spectral range (such as BD Horizon V500 and AmCyan), the most accurate compensation can be obtained using single stained cellular controls. Due to spectral differences between cells and beads in this channel, using BD™ CompBeads can result in spillover errors for V500 and AmCyan reagents. Therefore, the use of BD CompBeads or BD CompBeads Plus to determine spillover values for these reagents is not recommended. Different V500 reagents (eg, CD4 vs. CD45) can have slightly different fluorescence spillover therefore, it may also be necessary to use clone specific compensation controls when using these reagents.



**Analysis of CD8 on human lymphocytes.** Whole blood was stained with BD Horizon™ V500 Mouse Anti-Human CD8 and compared to whole blood stained with BD Horizon™ V500 Mouse IgG1, κ Isotype Control (clone X40, Cat. No. 560787). The isotype control is represented by a dashed line and the V500 Mouse Anti-Human CD8 by the solid line. Lymphocytes were selected by light scatter profile. Flow cytometry was performed on a BD FACSCanto™ II flow cytometry system.

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560774 Rev. 2



## 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™ V500 under optimum conditions, and unreacted BD Horizon™ V500 was removed.

## Application Notes

### Application

Flow cytometry	Routinely Tested
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## Suggested Companion Products

Catalog Number	Name	Size	Clone
560787	V500 Mouse IgG1, κ Isotype Control	0.1 mg	X40

## Product Notices

1. This reagent has been pre-diluted for use at the recommended Volume per Test. We typically use  $1 \times 10^6$  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. BD Horizon™ V500 has a maximum absorption of 415 nm and maximum emission of 500 nm. Before staining with this reagent, please confirm that your flow cytometer is capable of exciting the fluorochrome and discriminating the resulting fluorescence.
5. Species testing during development may have been performed with a different format of the same clone. Selected applications have been tested for cross-reactivity.
6. For fluorochrome spectra and suitable instrument settings, please refer to our Multicolor Flow Cytometry web page at [www.bdbiosciences.com/colors](http://www.bdbiosciences.com/colors).
7. Please refer to [www.bdbiosciences.com/pharming/protocols](http://www.bdbiosciences.com/pharming/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)

Schlossman SF, Boumsell L, Gilks W, et al, ed. *Leucocyte Typing V: White Cell Differentiation Antigens*. Oxford: Oxford University Press; 1995. (Clone-specific)

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560774 Rev. 2

