

Technical Data Sheet

V500 Syrian Hamster anti-Mouse CD3e**Product Information**

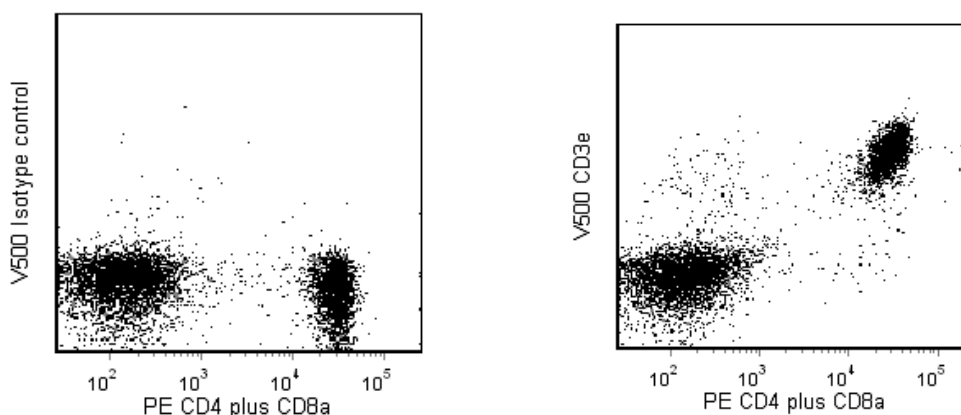
Material Number:	560773
Alternate Name:	CD3ε chain
Size:	25 µg
Concentration:	0.2 mg/ml
Clone:	500A2
Immunogen:	Mouse T-cell receptor
Isotype:	Syrian Hamster IgG2, κ
Reactivity:	QC tested: Mouse
Storage Buffer:	Aqueous buffered solution containing protein stabilizer, glycerol and ≤0.09% sodium azide.

Description

This antibody has been reported to react with the 25-kDa ε chain of the T-cell receptor-associated CD3 complex expressed on thymocytes, mature T lymphocytes, and NK-T cells on many mouse stains. Plate-bound and soluble hamster anti-mouse CD3e (clone 500A2) antibody has been reported to activate unprimed T cells in vitro. Activation of a mouse T-cell clone by soluble hamster anti-mouse CD3e (clone 500A2) can be blocked by Fab fragments of anti-CD4 antibody GK1.5, suggesting that the hamster anti-mouse CD3e (clone 500A2) antibody may bind an epitope of CD3e close to a site at which CD4 associates with the T-cell receptor. This hamster mAb to a mouse leukocyte antigen has been reported not to cross-react with rat leukocytes.

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.

When compensating dyes in this spectral range (such as 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 (e.g. 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.



CD3e expression in spleen. Splenocytes from BALB/c mice were simultaneously stained with PE rat anti-mouse CD4 mAb RM4-5 (Cat. no. 553048), PE rat anti-mouse CD8a mAb 53-6.7 (Cat. no. 553033), and Horizon™ V500 Hamster IgG2, λ Isotype Control (Cat. no. 560785, left panel) or Horizon™ V500 Hamster anti-Mouse CD3e (right panel) in the presence of BD Fc Block™ (Cat. no. 553142). Flow cytometry was performed on a BD FACSCanto™ II flow cytometry system.

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Preparation and Storage

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.

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

<u>Catalog Number</u>	<u>Name</u>	<u>Size</u>	<u>Clone</u>
560785	V500 Hamster IgG2, κ Isotype Control	0.1 mg	B81-3
553142	Purified Rat Anti-Mouse CD16/CD32 (Mouse BD Fc Block™)	0.5 mg	2.4G2
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5
553033	PE Rat Anti-Mouse CD8a	0.2 mg	53-6.7

Product Notices

1. Please refer to wwwbdbiosciences.com/pharming/en/protocols for technical protocols.
2. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
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. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at wwwbdbiosciences.com/colors.

References

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- Havran WL, Poenie M, Kimura J, Tsien R, Weiss A, Allison JP. Expression and function of the CD3-antigen receptor on murine CD4+8+ thymocytes. *Nature*. 1987; 330(6144):170-173. (Clone-specific)
- Kubo RT, Born W, Kappler JW, Marrack P, Pigeon M. Characterization of a monoclonal antibody which detects all murine alpha beta T cell receptors. *J Immunol*. 1989; 142(8):2736-2742. (Methodology: Flow cytometry)
- Ortaldo JR, Winkler-Pickett R, Mason AT, Mason LH. The Ly-49 family: regulation of cytotoxicity and cytokine production in murine CD3+ cells. *J Immunol*. 1998; 160(1):1158-1165. (Clone-specific)
- Portoles P, Rojo J, Golby A, et al. Monoclonal antibodies to murine CD3 epsilon define distinct epitopes, one of which may interact with CD4 during T cell activation. *J Immunol*. 1989; 142(12):4169-4175. (Clone-specific)