

Technical Data Sheet

Purified Hamster Anti-Mouse CD3e

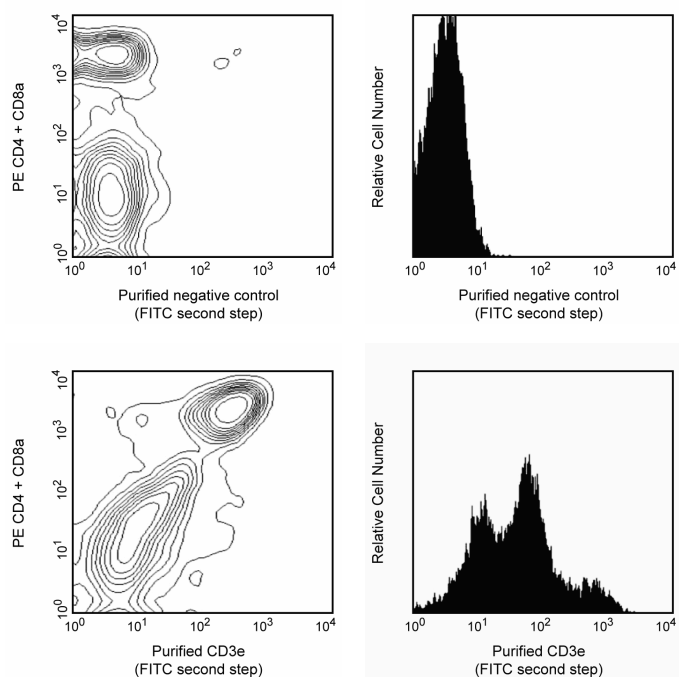
Product Information

Material Number:	553238
Alternate Name:	CD3ε chain
Size:	0.5 mg
Concentration:	0.5 mg/ml
Clone:	500A2
Immunogen:	Mouse T-cell receptor
Isotype:	Syrian Hamster IgG2, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤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.

This antibody is routinely tested by flow cytometric analysis. Other applications were tested at BD Biosciences Pharmingen during antibody development only or reported in the literature.



CD3e expression in spleen and thymus. BALB/c splenocytes were simultaneously stained with PE-conjugated anti-mouse CD4 mAb RM4-5 (Cat. No. 553048/553049, left panels), PE-conjugated anti-mouse CD8a mAb 53-6.7 (Cat. No. 553032/553033, left panels), and purified mAb 500A2 (bottom left panel), followed by FITC-conjugated anti-hamster IgG mAb G192-1 (Cat. No. 554026, left panels). BALB/c thymocytes were stained with purified 500A2 (bottom right panel) followed by FITC-conjugated G192-1 mAb (right panels). Flow cytometry was performed on a BD FACScan™ flow cytometry system.

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography. Store undiluted at 4° C.

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Application Notes

Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunoprecipitation	Reported
(Co)-stimulation	Reported

Recommended Assay Procedure:

It has been observed that pre-incubation of thymus cell suspensions at 37°C for 2 to 4 hours prior to staining enhances the ability of anti-CD3e and anti- $\alpha\beta$ TCR mAbs to detect the T-cell receptor on immature thymocytes. For IHC, we recommend the use of purified 500A2 mAb in our special formulation for immunohistochemistry, Cat. No. 550277. For activation of mouse T lymphocytes by plate-bound anti-CD3e mAb, we recommend BD Biosciences Biocoat™ 96-Well Clear Anti-Mouse CD3 T-Cell Activation Plates, Cat. No. 354720.

Suggested Companion Products

Catalog Number	Name	Size	Clone
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5
553032	PE Rat Anti-Mouse CD8a	0.1 mg	53-6.7
554026	FITC Mouse Anti-Hamster IgG	0.5 mg	G192-1
559277	Purified Hamster IgG2, κ Isotype Control	0.5 mg	B81-3

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharming/en/protocols for technical protocols.
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. Sodium azide is a reversible inhibitor of oxidative metabolism; therefore, antibody preparations containing this preservative agent must not be used in cell cultures nor injected into animals. Sodium azide may be removed by washing stained cells or plate-bound antibody or dialyzing soluble antibody in sodium azide-free buffer. Since endotoxin may also affect the results of functional studies, we recommend the NA/LE™ (No Azide/Low Endotoxin) antibody format, if available, for in vitro and in vivo use.
5. Although hamster immunoglobulin isotypes have not been well defined, BD Biosciences Pharmingen has grouped Armenian and Syrian hamster IgG monoclonal antibodies according to their reactivity with a panel of mouse anti-hamster IgG mAbs. A table of the hamster IgG groups, Reactivity of Mouse Anti-Hamster Ig mAbs, may be viewed at http://www.bdbiosciences.com/pharming/en/hamster_chart_11x17.pdf.

References

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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.(Biology)