Technical Data Sheet

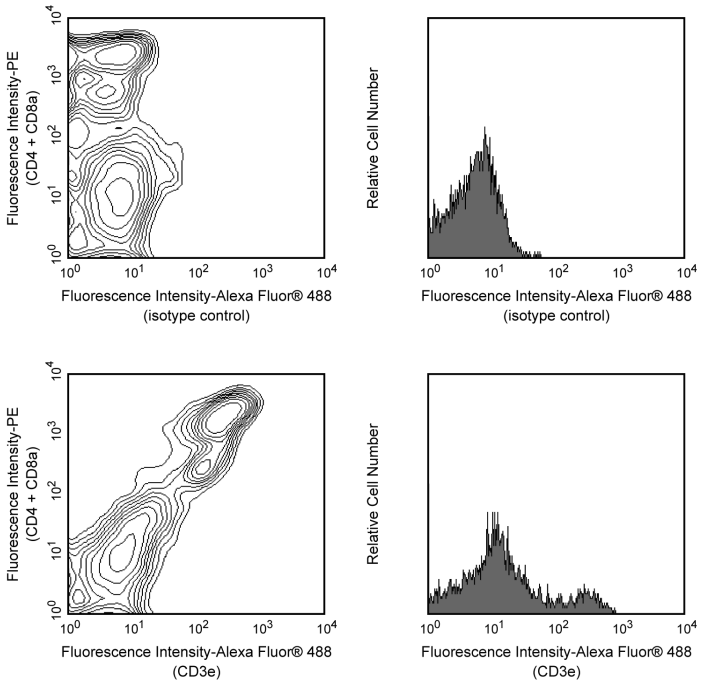
Alexa Fluor® 488 Hamster Anti-Mouse CD3e

Product Information

Material Number:	**557666**
Alternate Name:	CD3ε chain
Size:	0.1 mg
Concentration:	0.2 mg/ml
Clone:	145-2C11
Immunogen:	H-2Kb specific cytotoxic T lymphocyte clone BM10-37
Isotype:	Armenian Hamster IgG1, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

Description

The 145-2C11 antibody reacts with the 25-kDa ε chain of the T-cell receptor-associated CD3 complex, which is expressed on thymocytes, mature T lymphocytes, and NK-T cells. The cytoplasmic domain of CD3e participates in the signal transduction events which activate several cellular biochemical pathways as a result of antigen recognition. Soluble 145-2C11 antibody can activate either unprimed (naive) or primed (memory/preactivated) T cells in vivo or in vitro, in the presence of Fc receptor-bearing accessory cells. In contrast, plate-bound 145-2C11 can activate T cells in the absence of accessory cells. Soluble 145-2C11 antibody has been reported to induce re-directed lysis of Fc receptor-bearing target cells by CTL clones and can also block lysis of specific target cells by antigen-specific CTL's. Under some conditions, T-cell activation by 145-2C11 antibody has been reported to result in apoptotic cell death. The 145-2C11 antibody does not cross-react with rat leukocytes and it has been reported that pre-incubation of thymus cell suspensions at 37°C for 2-4 hours prior to staining enhances the ability of anti-CD3ε and anti-αβ TCR mAbs to detect the T-cell receptor on immature thymocytes.



CD3e expression in spleen and thymus. *C57BL/6 splenocytes were simultaneously stained with PE-conjugated anti-mouse CD8a mAb R4-5 (Cat. No. 553048), PE-conjugated anti-mouse CD8a mAb 53-6.7 (Cat. No. 553032) and either Alexa Fluor® 488-conjugated hamster IgG1κ isotype control mAb A19-3 (Cat. No. 557674) (top left panel) or Alexa Fluor® 488-conjugated mAb 145-2C11 (bottom left panel). C57BL/6 thymocytes were also stained with either Alexa Fluor® 488-conjugated isotype control (top right panel) or Alexa Fluor® 488-conjugated mAb 145-2C11 (bottom right panel). Flow cytometry was performed on a BD FACSCalibur™ flow cytometry 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 to Alexa Fluor® 488 under optimum conditions, and unreacted Alexa Fluor® 488 was removed.

Application Notes

Application

Flow cytometry	Routinely Tested

BD Biosciences

bdbiosciences.com

United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	888.259.0187	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

For country-specific contact information, visit **bdbiosciences.com/how_to_order/**

Suggested Companion Products

Catalog Number	Name	Size	Clone
557674	Alexa Fluor® 488 Hamster IgG1, κ Isotype Control	0.1 mg	A19-3
553048	PE Rat Anti-Mouse CD4	0.1 mg	RM4-5
553032	PE Rat Anti-Mouse CD8a	0.1 mg	53-6.7

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Alexa Fluor® 488 fluorochrome emission is collected at the same instrument settings as for fluorescein isothiocyanate (FITC).
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. The Alexa Fluor®, Pacific Blue™, and Cascade Blue® dye antibody conjugates in this product are sold under license from Molecular Probes, Inc. for research use only, excluding use in combination with microarrays, or as analyte specific reagents. The Alexa Fluor® dyes (except for Alexa Fluor® 430), Pacific Blue™ dye, and Cascade Blue® dye are covered by pending and issued patents.
5. Alexa Fluor is a registered trademark of Molecular Probes, Inc., Eugene, OR.
6. 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/pharmingen/hamster_chart_11x17.pdf.
7. For fluorochrome spectra and suitable instrument settings, please refer to our Fluorochrome Web Page at www.bdbiosciences.com/colors.
8. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

Duke RC, Cohen JJ, Boehme SA, et al. Morphological, biochemical, and flow cytometric assays of apoptosis. In: Coligan J, Kruisbeek AM, Margulies D, Shevach EM, Strober W, ed. *Current Protocols in Immunology*. New York: John Wiley and Sons; 1995:3.17.1-3.17.33.(Methodology: Activation, Apoptosis, Immunoprecipitation)

Isakov N, Wange RL, Burgess WH, Watts JD, Aebersold R, Samelson LE. ZAP-70 binding specificity to T cell receptor tyrosine-based activation motifs: the tandem SH2 domains of ZAP-70 bind distinct tyrosine-based activation motifs with varying affinity. *J Exp Med.* 1995; 181(1):375-380.(Biology: Immunoprecipitation)

Kruisbeek AM, Shevach EM. Proliferative assays for T cell function. In: Coligan J, Kruisbeek AM, Margulies D, Shevach EM, Strober W, ed. *Current Protocols in Immunology.* New York: John Wiley and Sons; 1991:3.12.1-3.12.14.(Methodology: Activation, Stimulation)

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.(Biology)

Leo O, Foo M, Sachs DH, Samelson LE, Bluestone JA. Identification of a monoclonal antibody specific for a murine T3 polypeptide. *Proc Natl Acad Sci U S A.* 1987; 84(5):1374-1378.(Immunogen: Activation, Blocking, Cytotoxicity, Immunoprecipitation, Stimulation)

Nakano H, Yamazaki T, Miyatake S, Nozaki N, Kikuchi A, Saito T. Specific interaction of topoisomerase II beta and the CD3 epsilon chain of the T cell receptor complex. *J Biol Chem.* 1996; 271(11):6483-6489.(Biology: Immunoprecipitation)

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: Activation, Immunoprecipitation, Stimulation)

Shinkai Y, Alt FW. CD3 epsilon-mediated signals rescue the development of CD4+CD8+ thymocytes in RAG-2-/- mice in the absence of TCR beta chain expression. *Int Immunol.* 1994; 6(7):995-1001.(Biology: Activation, Stimulation)