

CD3E Recombinant Monoclonal Antibody [BL-298-5D12]

Rabbit Recombinant Monoclonal

Purified	Protein ID	NP_000724.1
Catalog No. A700-016-T	GeneID	916
Lot No. A700-016-T-2		



APPLICATIONS	WB, IP, IHC, ICC, IHC-IF, F, mIF
SPECIES REACTIVITY	Human
AMOUNT	10 µl (5+ tests)
CONCENTRATION	50 µg/ml
STORAGE/SHELF LIFE	2 – 8° C / 1 year from date of receipt
PHYSICAL STATE	Liquid
BUFFER	Borate Buffered Saline (BBS) pH 8.2 with 0.1% BSA and 0.09% Sodium Azide
ISOTYPE	IgG
CLONE #	BL-298-5D12
ORIGIN	USA
PRODUCTION PROCEDURES	Recombinant antibody was purified from cell culture supernatant. Immunogen was a synthetic peptide representing a region of CD3E and maps to a region between residues 157 to 207 of human CD3e Molecule, Epsilon (CD3-TCR Complex) according to the numbering given in entry NP_000724.1 (GeneID 916).

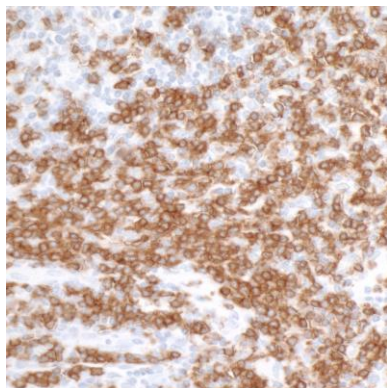
APPLICATIONS Centrifuge tube to remove product from lid. Optimal working dilutions should be determined experimentally by the investigator. Prepare working dilution immediately before use.

Western Blot	1:1000
Immunoprecipitation	20 µl/mg lysate
Immunohistochemistry	1:100 – 1:500. Epitope retrieval with citrate buffer pH 6.0 is recommended for FFPE tissue sections.
Immunocytochemistry	1:100 – 1:500. Epitope retrieval with citrate buffer pH6.0 is recommended for FFPE cell sections.
Immunofluorescence (IHC)	1:100 to 1:500. Epitope retrieval with citrate buffer pH6.0 is recommended for FFPE cell sections.
Flow Cytometry	1 µl per 1 X 10 ⁶ cells
Multiplex Immunofluorescence	1:250

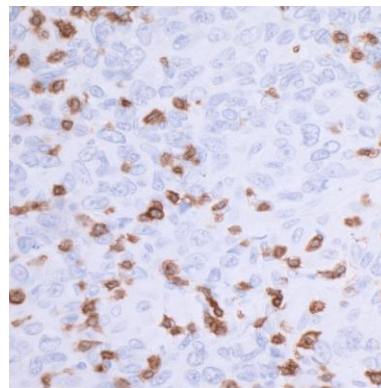
IHC HUMAN CONTROLS Appendix, Metastatic Lymph Node, Tonsil, Jurkat Cells, MOLT-4 Cells

ADDITIONAL INFO <https://www.bethyl.com/product/A700-016-T>
Use the link above to view SDS, a current list of citations, and other product specific information.

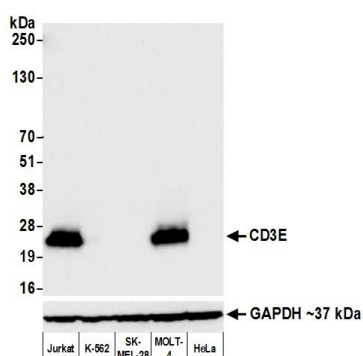
This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc.
Eric McIntush, PhD | Chief Scientific Officer Date: July 26, 2019



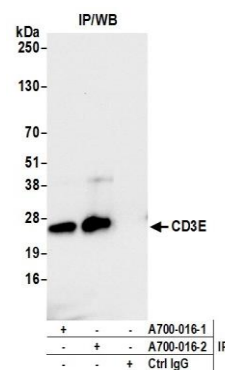
Detection of human CD3E in FFPE tonsil by IHC. *Antibody:* Rabbit anti-CD3E recombinant monoclonal [BL-298-5D12] (A700-016 lot 2). *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-501P). *Substrate:* DAB.



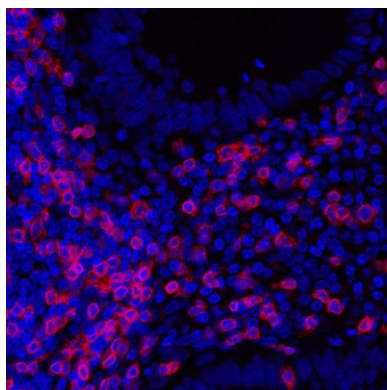
Detection of human CD3E in FFPE breast carcinoma by IHC. *Antibody:* Rabbit anti-CD3E recombinant monoclonal [BL-298-5D12] (A700-016 lot 2). *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-501P). *Substrate:* DAB.



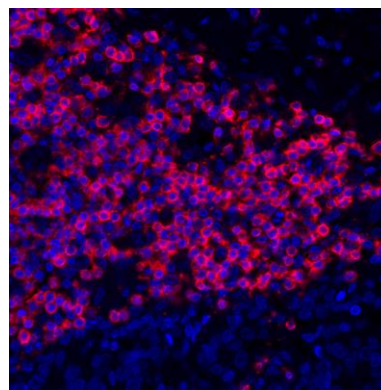
Detection of human CD3E by western blot. *Samples:* Whole cell lysate (50 µg) from Jurkat, K-562, SK-MEL-28, MOLT-4, and HeLa cells prepared using NETN lysis buffer. *Antibody:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 2) used at 1:1000. *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-101P). Chemiluminescence with an exposure time of 3 seconds. Lower Panel: Rabbit anti-GAPDH (A300-639A).



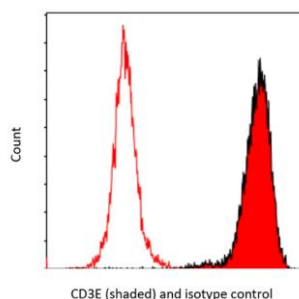
Detection of human CD3E by western blot of immunoprecipitates. *Samples:* Whole cell lysate (1.0 mg per IP reaction; 20% of IP loaded) from Jurkat cells prepared using NETN lysis buffer. *Antibodies:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 2) used for IP at 20 µl/mg lysate. CD3E was also immunoprecipitated by a previous lot of this antibody (lot A700-016-1). For blotting immunoprecipitated CD3E, A700-016 was used at 1:1000. Chemiluminescence with an exposure time of 3 seconds.

**Detection of human CD3E by immunohistochemistry.**

Sample: FFPE section of appendix. *Antibody:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 1) used at 1:250. *Secondary:* DyLight® 594-conjugated goat anti-rabbit IgG (A120-101D4).

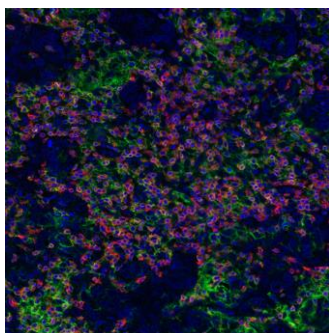
**Detection of human CD3E (red) by immunohistochemistry.**

Sample: FFPE section of human lung carcinoma. *Antibody:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016-T lot 1) used at 1:250. *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-501P). *Substrate:* Opal™. *Counterstain:* DAPI (blue).



CD3E (shaded) and isotype control

Detection of human CD3E by flow cytometry. *Sample:* Jurkat cells were fixed in 4% formaldehyde and permeabilized with 90% methanol. *Antibody:* 1E6 cells were incubated with 1 ul of rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016-T lot 1) (red shaded) or isotype control (unshaded). *Secondary:* DyLight® 650-conjugated goat anti-rabbit IgG (A120-201D5).



Detection of human CD3E (orange), CD8 alpha (red), and PD-L1 (green) in FFPE breast carcinoma by IHC-IF. Rabbit anti-CD3E recombinant monoclonal [BL-298-5D12] (A700-016), rabbit anti-CD8 alpha recombinant monoclonal [BLR044F] (A700-044), rabbit anti-PD-L1 recombinant monoclonal [BLR020E] (A700-020). *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-501P). *Substrate:* Opal™ 520, 620, and 690. *Counterstain:* DAPI (blue).

