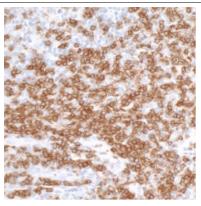
CD3E Recombinant Monoclonal Antibody [BL-298-5D12]

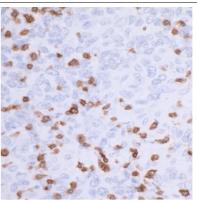
Rabbit Recombinant Monoclonal					C C M	
Purified		Р	Protein ID	NP_000724.1		
Catalog No.	-		GenelD	916	RETHVI	
Lot No.	A700-	016-T-2			LABORATORIES, INC	
APPLICATIONS		WB, IP, IHC, ICC, IHC–IF, F, mIF				
SPECIES REACTIVITY		Human				
AMOUNT		10 µl (5+ tests)				
CONCENTRATION		50 μg/ml				
STORAGE/SHELF 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	1:1	000		
		Immunoprecipitat	tion 20	µl/mg lysate		
		Immunohistocher	,	00 – 1:500. Epitope retrieval with citrate ommended for FFPE tissue sections.	buffer pH 6.0 is	
		Immunocytochem		00 – 1:500. Epitope retrieval with citrate ommended for FFPE cell sections.	buffer pH6.0 is	
		Immunofluoresce (IHC)		00 to 1:500. Epitope retrieval with citrate ommended for FFPE cell sections.	e buffer pH6.0 is	
		Flow Cytometry	lμ	l per 1 X 10^6 cells		
		Multiplex Immunofluoresce	1:2 ence	50		
IHC HUMAN CONTROLS Append		Appendix, Metasta	endix, 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

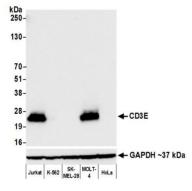
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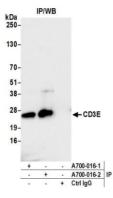


Detection of human CD3E in FFPE tonsil by IHC. *Antibody:* Rabbit anti-CD3E recombinant monoclonal [BL-298-5D12] (A700-016 lot 2). *Secondary:* HRP-conjugated goat antirabbit IgG (A120-501P). *Substrate:* DAB.



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



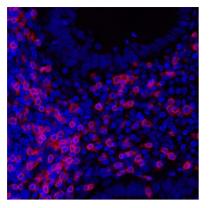


Detection of human CD3E by western blot. Samples: Whole cell lysate (50 µg) from Jurkat, K-562, SK-MEL-28, MOLT-4, and HeLa cells prepared using NETN lysis buffer. Antibody: Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 2) used at 1:1000. Secondary: HRP-conjugated goat 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

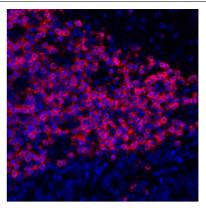
immunoprecipitates. *Samples:* Whole cell lysate (1.0 mg per IP reaction; 20% of IP loaded) from Jurkat cells prepared using NETN lysis buffer. *Antibodies:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 2) used for IP at 20 µl/mg lysate. CD3E was also immunoprecipitated 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.

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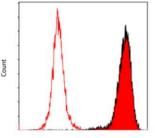


Detection of human CD3E by immunhistochemistry. *Sample:* FFPE section of appendix. *Antibody:* Rabbit anti-CD3E recombinant monoclonal antibody [BL-298-5D12] (A700-016 lot 1) used at 1:250. *Secondary:* DyLight® 594-conjugated goat anti-rabbit IgG (A120-101D4).



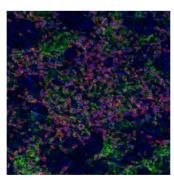
Detection of human CD3E (red) by

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 antirabbit 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).

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4

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