

CD247/CD3Z Recombinant Monoclonal Antibody [BL-336-1B2]

Rabbit Recombinant Monoclonal

Purified		RefSeq ID	NP_000725.1
Catalog No.	A700-017-T	Uniprot ID	P20963
Lot No.	4	GeneID	919

APPLICATIONS	WB, IP, IHC, IHC-IF, Flow Cyt, SW-Size
SPECIES REACTIVITY	Human
PRESUMED REACTIVITY	Based on 100% sequence identity, this antibody is predicted to react with Mouse, Goat and Pig
AMOUNT	10 µl (5+ tests)
CONCENTRATION	100 µg/ml
STORAGE/SHELF LIFE	2 – 8°C / 1 year from date of receipt
PHYSICAL STATE	Liquid
BUFFER	Phosphate Buffered Saline (PBS) with 0.1% BSA and 0.09% Sodium Azide
ISOTYPE	IgG
CLONE #	BL-336-1B2
ORIGIN	USA
PRODUCTION PROCEDURES	Recombinant antibody was purified from cell culture supernatant.

Immunogen was a synthetic peptide representing a region between residue 150 and the C-terminus (residue 164) of human T-cell surface glycoprotein CD3 zeta chain using the numbering given in entry NP_932170.1 (GeneID 919).

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.
Immunofluorescence (IHC)	1:100 – 1:500. Epitope retrieval with citrate buffer pH 6.0 is recommended for FFPE tissue sections.
Flow Cytometry	Fixed in 4% formaldehyde and permeabilized with 90% methanol. 0.5 µl per 1 x 10 ⁶ cells.
Simple Western™-Size	1:10 – 1:250

APPLICATION NOTES	All western blot analysis is performed using 5% Milk-TBST for blocking and as antibody diluent. Primary antibody is incubated overnight.
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Western blots of cell lysates are performed using Goat anti-Rabbit IgG Heavy and Light Chain Antibody (A120-101P).

Western blots of immunoprecipitates are performed using Goat anti-Rabbit Light Chain HRP Conjugate (A120-113P) with 5% Normal Pig Serum (S100-020) added to the blocking buffer.

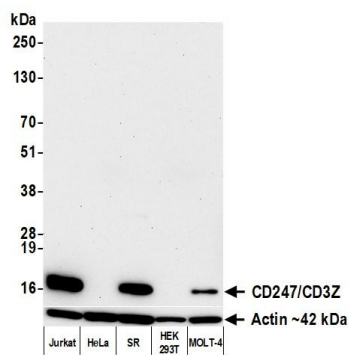
IHC HUMAN CONTROLS	Appendix, Metastatic Lymph Node, Tonsil
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ADDITIONAL INFO	https://www.fortislifesciences.com/p/A700-017-T
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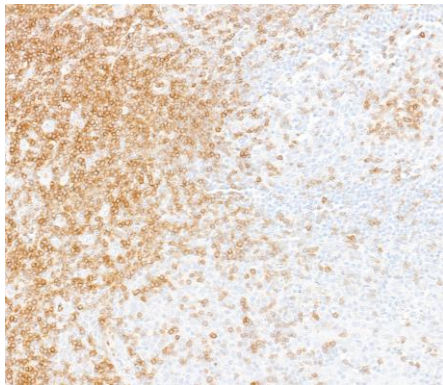
Use the link above to view SDS, a current list of citations, and other product specific information.

This document certifies that this product has met all of the quality control standards defined by Bethyl Laboratories, Inc.
Michael Spencer, PhD Date: August 19, 2024

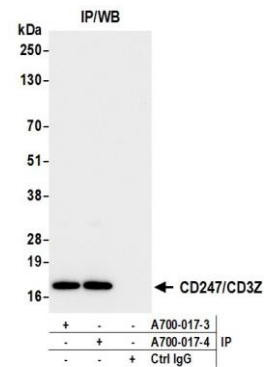
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**Detection of human CD247/CD3Z by western blot.**

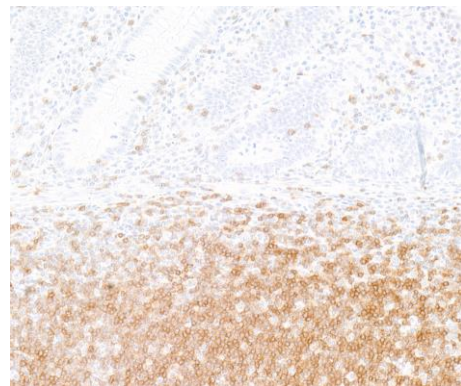
Samples: Whole cell lysate (10 µg) from Jurkat, HeLa, SR, HEK293T, and MOLT-4 cells prepared using NETN lysis buffer. **Antibody:** Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017) used at 1:1000. **Secondary:** HRP-conjugated goat anti-rabbit IgG (A120-101P). **Detection:** Chemiluminescence with an exposure time of 3 minutes. Lower Panel: Rabbit anti-Actin recombinant monoclonal antibody [BLR057F] (A700-057).



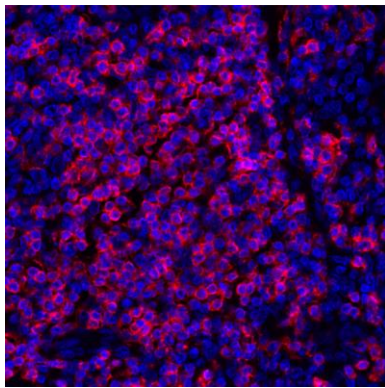
Detection of human CD247/CD3Z by immunohistochemistry. **Sample:** FFPE section of tonsil. **Antibody:** Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017). **Secondary:** HRP-conjugated goat anti-rabbit IgG (A120-501P).

**Detection of human CD247/CD3Z by western blot of immunoprecipitates.**

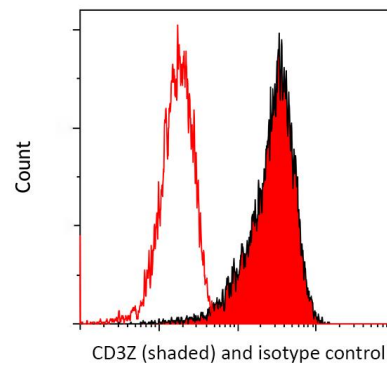
Samples: Whole cell lysate (1.0 mg per IP reaction; 10% of IP loaded) from Jurkat cells prepared using NETN lysis buffer. **Antibodies:** Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017) used for IP at 20 µl/mg lysate. CD247/CD3Z was also immunoprecipitated by a previous lot of this antibody. For blotting immunoprecipitated CD247/CD3Z, A700-017 was used at 1:1000. **Detection:** Chemiluminescence with an exposure time of 3 seconds.



Detection of human CD247/CD3Z by immunohistochemistry. **Sample:** FFPE section of appendix. **Antibody:** Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017). **Secondary:** HRP-conjugated goat anti-rabbit IgG (A120-501P).



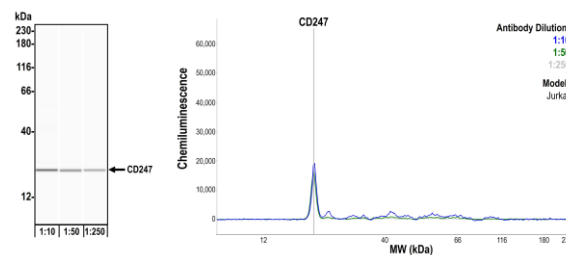
Detection of human CD247/CD3Z (red) by immunohistochemistry. *Sample:* FFPE section of human tonsil. *Antibody:* Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017) used at 1:250. *Secondary:* HRP-conjugated goat anti-rabbit IgG (A120-501P). *Substrate:* Opal™. *Counterstain:* DAPI (blue).



CD3Z (shaded) and isotype control

Detection of human CD247/CD3Z by flow cytometry.

Sample: Jurkat cells were fixed in 4% formaldehyde and permeabilized with 90% methanol. *Antibody:* 1E6 cells were incubated with 0.5 ul of rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017) (red shaded) or isotype control (unshaded). *Secondary:* DyLight® 650-conjugated goat anti-rabbit IgG (A120-201D5).



Detection of human CD247/CD3Z by Simple Western.

Samples: Whole cell lysate (2.0 mg/mL) from Jurkat cells prepared using NETN lysis buffer. *Antibody:* Rabbit anti-CD247/CD3Z recombinant monoclonal antibody [BL-336-1B2] (A700-017) used at 1:10, 1:50, 1:250. *Separation and Detection:* SallySue ProteinSimple instrument with the 12-230 kDa separation module and anti-Rabbit detection module. Left Panel: Virtual Lane View. Right Panel: Electropherogram.