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Anti-Hu CD3 zeta (pY153) Purified CD247 Monoclonal Antibody

Catalog Number: M02421-1

About CD247

CD3 complex is crucial in transducing antigen-recognition signals into the cytoplasm of T cells and in regulating the cell surface expression of the TCR complex. T cell activation through the antigen receptor (TCR) involves the cytoplasmic tails of the CD3 subunits CD3 gamma, CD3 delta, CD3 epsilon and CD3 zeta (CD247). These CD3 subunits are structurally related members of the immunoglobulins super family encoded by closely linked genes on human chromosome 11. The CD3 components have long cytoplasmic tails that associate with cytoplasmic signal transduction molecules. This association is mediated at least in part by a double tyrosine-based motif present in a single copy in the CD3 subunits. CD3 may play a role in TCR-induced growth arrest, cell survival and proliferation.

Overview

Product Name	Anti-Hu CD3 zeta (pY153) Purified CD247 Monoclonal Antibody
Reactive Species	Human, Mouse
Description	Boster Bio Anti-Hu CD3 zeta (pY153) Purified CD247 Monoclonal Antibody (Catalog# M02421-1). Tested in Flow Cytometry, WB application(s). This antibody reacts with Human, Mouse.
Application	Flow Cytometry, WB
Clonality	Monoclonal EM-17
Formulation	Phosphate buffered saline (PBS), pH 7.4, 15 mM sodium azide
Storage Instructions	Store at 2-8°C. Do not freeze.
Host	Mouse
Uniprot ID	P20963

Technical Details

Immunogen	A phospho specific peptide corresponding to the amino acids surrounding tyrosine 153 of mouse CD3 zeta linked to KLH. The mouse monoclonal antibody EM-17 recognizes phosphorylated intracellular tyrosine 153 of CD3 zeta chain (CD247), which is a component of TCR/CD3 complex expressed on T cells.
Predicted Reactive Species	Primate
Cross Reactivity	This antibody does not cross-react with Thy-1.1 alloantigen.
Isotype	Mouse IgG1
Form	Liquid
Concentration	1 mg/ml

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Purification	Purified by protein-A affinity chromatography.
Suggested Dilutions	Western blotting: 2 - 5 ug/ml; positive control: Jurkat cells lysate treated with pervanadate, splenocyte lysate of Balb/c or F1 mouse treated with pervanadate, non-reducing conditions recommended. Flow cytometry: Intracellular staining; recommended dilution: 1-4 ug/ml; positive control: Jurkat cells treated with pervanadate.

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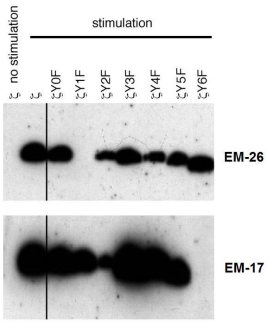
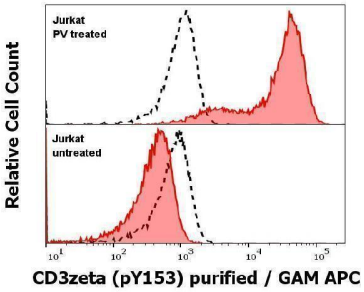
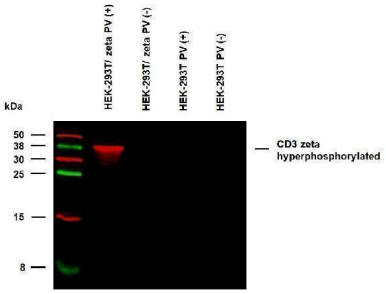
Anti-Hu CD3 zeta (pY153) Purified CD247 Monoclonal Antibody (M02421-1) Images



Western blotting analysis using monoclonal antibodies EM-26 (anti-CD3 zeta phospho-Tyr72) and EM-17 (anti-CD3 zeta phospho-Tyr153) reacting with particular phosphorylated human CD3 zeta mutants. The Y1F and Y6F mutatants lack phosphotyrosine 72 and 153, respectively.



Anti-Hu CD3 zeta (pY153) purified antibody (clone EM-17) works in Flow Cytometry application Analysis of the antibody staining was performed on Jurkat cells treated or untreated with pervanadate (PV) prior to the fixation and permeabilization of cell suspension with cold methanol. Anti-Hu CD3 zeta (pY153) purified antibody (concentration in sample 1 µg/ml, GAM APC, red-filled histogram) binds specifically to phosphorylated tyrosine 153 (pY153) of CD3 zeta chain in PV treated, methanol permeabilized Jurkat cells (upper panel), but not to untreated methanol permeabilized control cells (lower panel). Level of non-specific binding was assessed using Mouse IgG1 isotype control purified (MOPC-21) under same conditions (concentration in sample 1 µg/ml, GAM APC, black-dashed histogram).



Anti-Hu CD3 zeta (pY153) Purified (clone EM-17) specificity verification by WB. The specificity of EM-17 antibody to phosphorylated Tyr 153 (CD3 zeta chain) was assessed by analysis of binding signals in HEK293T transfected with CD3 zeta/ZAP-70 construct followed by pervanadate (PV) treatment in comparison to the series of control cells – PV untreated transfectants, and both PV treated and untreated mock HEK293T cells. Western blotting analysis was performed on whole cell extracts (RIPA lysis buffer with PhosSTOP and pervanadate), mixed and heated (100°C, 5 min) with non-reducing SDS-loading buffer. Samples were resolved using 15% SDS-PAGE gel. Nitrocellulose membrane blot was probed with mouse IgG1 monoclonal antibody EM-17 (1 µg/ml). Subclass-specific secondary antibody IRDye 680LT Goat-anti-Mouse IgG (red) was used for fluorescent Western blot detection.

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