Technical Data Sheet

Purified Mouse Anti-Zap70 (pY493)

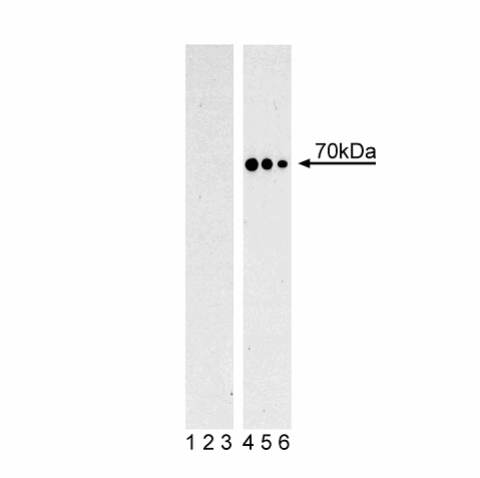
Product Information

Material Number:	**558247**
Size:	0.1 mg
Concentration:	0.5 mg/ml
Clone:	1a/Zap70 (pY493)
Isotype:	Mouse IgG1, κ
Reactivity:	QC Testing: Human Tested in Development: Mouse
Target MW:	70 kDa
Storage Buffer:	Aqueous buffered solution containing ≤0.09% sodium azide.

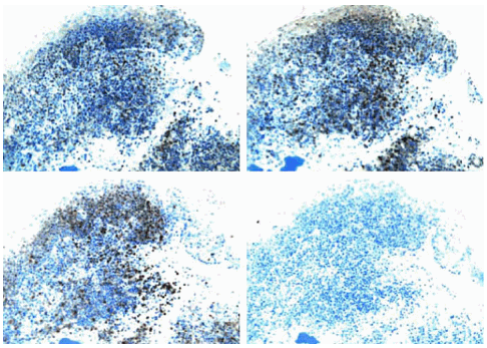
Description

The 70-kDa ζ chain-associated protein (ZAP70) is a protein tyrosine kinase (PTK) that associates with the ζ subunit of the T cell antigen receptor (TCR) and undergoes tyrosine phosphorylation following TCR stimulation. ZAP70 contains two SH2-like domains with the PTK domain located at the C-terminus. It appears that both ZAP70 and Syk are recruited to the phosphorylated CD3 and ζ subunits after TCR stimulation. TCR-mediated Lck activity leads to phosphorylation of ZAP70 on Tyrosine 493 (Y493) in the regulatory loop of the kinase domain leading to upregulation of ZAP70 kinase activity. The significance of ZAP70 activation in mediating TCR signal transduction has been confirmed by showing that ZAP70 activity is absent in an autosomal recessive form of severe combined immunodeficiency (SCID). This is due to mutations affecting the ZAP70 kinase domain which affect the stability of the protein and TCR signaling.

The 1a/Zap70 (pY493) antibody recognizes ZAP70 phosphorylated at Y493.



Western blot analysis of ZAP70 (pY493). *Lysates from control (left panel) and hydrogen peroxide-treated (right panel) Jurkat cells were probed with purified Mouse anti-ZAP70 (pY493) at concentrations of 8.0 (lanes 1,4), 4.0 (lanes 2,5), and 2.0 µg/ml (Lanes 3,6). ZAP70 (pY493) is identified as a strong band of 70 kDa in the treated cells.*



ZAP70 staining on tonsil. *Fresh human tonsil was incubated in 5 mM Pervanadate solution for 2 hours, then fixed in formalin and processed. Following antigen retrieval with BD Retrievagen A buffer (Cat. No. 550524), the sections were either left untreated (left column) or treated with a phosphatase to eliminate all phosphorylation (right column). The tissue sections were stained with either purified Mouse anti-ZAP-70 (Cat. No. 610239 or 610240, top row) or purified Mouse anti-ZAP70 (pY493) (bottom row) with Hematoxylin counterstaining. Original magnification: 20X.*

Preparation and Storage

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.
Store undiluted at 4°C.

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Application Notes

Application

Western blot	Routinely Tested
Immunohistochemistry-formalin (antigen retrieval required)	Tested During Development

Suggested Companion Products

Catalog Number	Name	Size	Clone
554002	HRP Goat Anti-Mouse Ig	1.0 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.
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.

References

Chan AC, Dalton M, Johnson R, et al. Activation of ZAP-70 kinase activity by phosphorylation of tyrosine 493 is required for lmphocyte antigen receptor function. *EMBO J.* 1995; 14:2499-2508.(Biology)

Chan AC, Kadlecek TA, Elder ME, et al. ZAP-70 deficiency in an autosomal recessive form of severe combined immunodeficiency. *Science.* 1994; 264:1599-1601.(Biology)

Mege D, Di Bartolo V, Germain V, Tuosto L, Michel F, Acuto O. Mutation of tyrosines 492/493 in the kinase domain of ZAP-70 affects multiple T-cell receptor signaling pathways. *J Biol Chem.* 1996; 271:32644-32652.(Biology)