

Product datasheet for AM03112PU-N

OriGene Technologies, Inc.

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ZAP70 Mouse Monoclonal Antibody [Clone ID: ZAP-03]

Product data:

Product Type: Primary Antibodies

Clone Name: ZAP-03
Applications: FC, IF, WB

Recommended Dilution: Flow Cytometry (2-5 µg/ml):Intracellular staining.

Positive control: HPB-ALL human peripheral blood T cell leukemia cell line.

Western Blotting (0,5 μg/ml): Under Reducing conditions. SDS-PAGE (10% separating gel).

Positive control: HPB-ALL human peripheral blood T cell leukemia cell line.

Negative control: RAMOS human Burkitt lymphoma cell line.

Sample preparation: Resuspend approx. 50 mil. cells in 1 ml cold lysis buffer (1% laurylmaltoside in 20 mM Tris/Cl, 100 mM NaCl pH 8.2, 50 mM NaF including Protease inhibitor Cocktail). Incubate 60 min on ice. Centrifuge to remove cell debris. Mix lysate (1:1)

with reducing Laemmli SDS-PAGE sample buffer. Boil for 5 min.

<u>Immunocytochemistry</u>.

Reactivity: Human
Host: Mouse
Isotype: IgG1

Clonality: Monoclonal

Immunogen: Bacterially expressed fusion protein representing C-terminal part (160 amino acids) of human

ZAP-70.

Specificity: The antibody ZAP-03 reacts with ZAP70, a 70 kDa protein tyrosine kinase expressed in T and

NK cells.

ZAP70 is a molecule susceptible to degradation. It is recommended to use freshly prepared cell lysates (protease inhibitors are essential) to avoid non-specific staining of

degradation products.

Formulation: PBS, pH 7.4 containing 15 mM sodium azide as preservative.

State: Aff - Purified

State: Liquid purified IgG fraction (> 95% pure by SDS-PAGE).

Concentration: lot specific

Purification: Protein A affinity chromatography.





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Conjugation: Unconjugated

Storage: Store the antibody undiluted at 2-8°C for one month or (in aliquots) at -20°C for longer.

Avoid repeated freezing and thawing.

Stability: Shelf life: one year from despatch.

Gene Name: zeta chain of T cell receptor associated protein kinase 70kDa

Database Link: Entrez Gene 7535 Human

P43403

Background: The ZAP70 (zeta-associated protein of 70 kDa) tyrosine kinase was identified as a tyrosine

phosphoprotein that associates with TCR zeta subunit and undergoes tyrosine

phosphorylation following TCR stimulation. ZAP70 is a Syk family tyrosine kinase primarily expressed in T and NK cells that plays an essential role in signaling through the TCR. TCR-mediated activation of T cells is crucial to the immune response. In humans, ZAP70 gene mutations resulting in lower ZAP70 protein expression levels or expression of catalytically inactive ZAP70 proteins, have been identified. ZAP70 deficiency results in the absence of mature CD8+ T cells and the prevention of TCR-mediated activation of CD4+ T cells, and it can lead to severe combined immunodeficiency. In patients with chronic lymphocytic leukemia (B-CLL), ZAP70 expression on B cell was shown to be correlated with disease progression and

survival.

ZAP70 is cytosolic protein migrating at 70 kDa in SDS-PAGE. It contains two N-terminal SH2 domains (Src homology domain 2) and a C-terminal kinase domain. Crystal structure of the ZAP70 SH2 domains in complex with a TCR zeta subunit peptide was described. During T cell activation , the binding of ZAP70 SH2 domains to the phosphorylated zeta subunit on the

activated TCR complex causes a colocalization with the Lck tyrosine kinase that

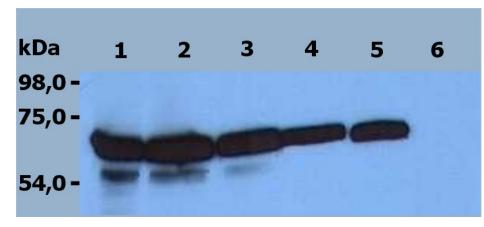
phosphorylates ZAP70 on Tyr493 in the activation loop. ZAP70 autophosphorylates multiple tyrosines in the region between the SH2 domains and the kinase domain, including the binding sites for additional SH2-containing signaling proteins such as SLP76, LAT, Lck, PLCgamma1, Vav, Shc, Ras-GAP, and Abl. ZAP70-mediated activation of these downstream effectors leads to the release of intracellular calcium stores, and the transcription of

interleukin-2 and other genes important for an immune response.

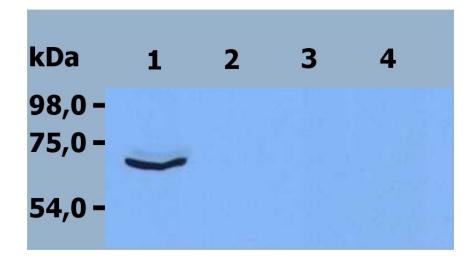
Synonyms: ZAP-70, ZAP 70, SRK



Product images:



Western Blotting analysis (reducing conditions) of HPB-ALL peripheral blood T cell leukemia cell line. Lane 1, 2, 3, 4: immunostaining with dilution range of anti-ZAP-70 (ZAP-03; 4 ug/ml (1), 2 ug/ml (2), 1 ug/ml (3), 0.5 ug/ml (4) Lane 5: immunostaining with anti-ZAP-70 comparative antibody Lane 6: immunostaining with Isotype mouse IgG1 control



Western Blotting analysis (reducing conditions) of HPB-ALL peripheral blood T cell leukemia cell line (1, 3) and RAMOS human Burkitt lymphoma cell line (2, 4); Lane 1, 2: immunostaining with anti-ZAP-70 (ZAP-03; 0.5 ug/ml) Lane 3, 4: immunostaining with Isotype mouse IgG1 control