



Antibody Specification Sheet

Anti-Human SLP-76 - Purified



Catalog reference: AS55-P

Size: 0.1 mg

Antibody Information:

Antigen: recombinant human SLP-76 fusion protein.

Ig Class: mIgG2a.

Form: purified.

Specificity: recognizes human SH2 (Src homology 2) domain-Leukocyte Protein of 76 kDa.

Antibody Source: monoclonal antibody from BALB/c-

Applications:

derived hybridoma SLP-76-H3.

Production: *in vitro* cell culture.

Purification: Protein A affinity chromatography.

Purity: ≥95%.

Formulation: provided as a 0.2 µm sterile-filtered solution in Ca⁺⁺ & Mg⁺⁺ free

Dulbecco's PBS with 0.1% sodium azide.

Concentration: 0.5 mg/ml.

Recommendations: Western Blot and Immunoprecipitation.

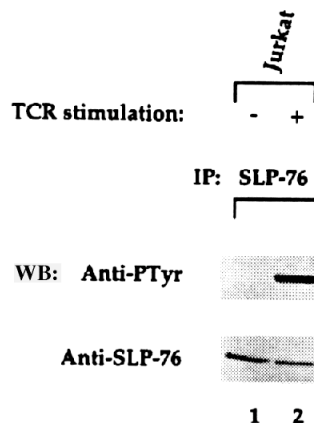
Storage conditions: store undiluted at 4 °C or in aliquots at ≤ -20 °C.

Western Blot: This antibody can be used at 1 µg/ml to visualize 1 x 10⁵ Jurkat cells per lane.

Immunoprecipitation: This antibody can be used at a range from 1 to 5 µg per 1 x 10⁶ Jurkat cells.

References: Wardenburg, J.B., Fu, C., Jackmann, J.K., Flotow, H., Wilkinson, S.E., Williams, D.H., Johnson, R., Kong, G., Chan, A.C., Findel, P.R. (1996). Phosphorylation of SLP-76 by the ZAP-70 protein-tyrosine kinase is required for T-cell receptor function. *J. Biol. Chem.* **271**, 19641-19644.

Jackman, K. J., Motto, D. G., Sun, Q., Tanemoto, M., Turk, C.W., Peltz, G. A., Koretzky, G. A., Findell, P.R. (1995). Molecular cloning of SLP-76, a 76-kDa tyrosine phosphoprotein associated with Grb2 in T cells. *J. Biol. Chem.* **270**, 7029-7032.



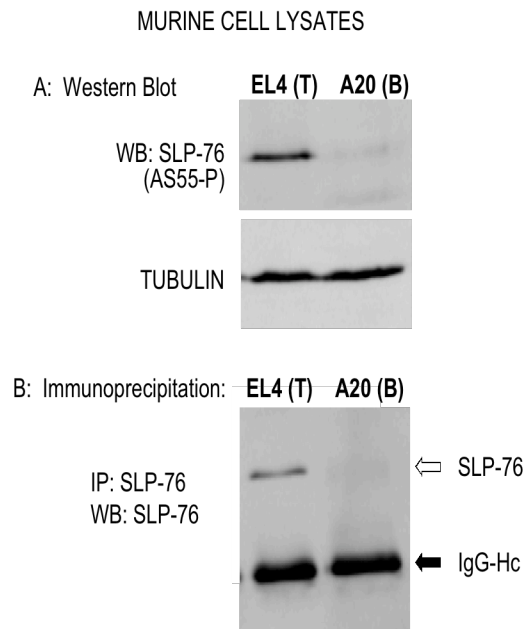
Immunoprecipitation (IP) by AS55 of SLP-76 from control and stimulated Jurkat T cells followed by western blotting (WB) with an anti-phosphotyrosine antibody or AS55 anti-SLP-76.

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For Western blot analysis of whole cell lysates (A), the anti-SLP-76 antibody was used at a final concentration of 1 ug/ml. Each lane was loaded with whole cell lysate derived from 4×10^6 cells. For immunoprecipitation (B), whole cell lysates were prepared from 7.5×10^6 cells and brought up to a final volume of 1 ml with PBS. Lysates were pre-cleared with Protein G-Sepharose beads, then incubated with 2 ug of the anti-SLP-76 antibody overnight at 4° C. Protein G-Sepharose beads were added for an additional 2 hours of incubation at 4° C. Beads were washed, boiled in SDS-reducing buffer, and eluted proteins were subjected to SDS-PAGE and Western blotting as described. EL4, murine T cell line; A20, murine B cell line.

General Information: SLP-76 is a substrate of the TCR-stimulated protein tyrosine kinase that function in the signal transduction cascade linking the TCR with IL-2 gene expression. Alignment of murine and human SLP-76 nucleic acid (80% identity) and predicted protein sequences (84% identity) indicates a very high degree of sequence homology. Human and murine SLP-76 mRNA demonstrate an identical pattern of tissue-specific expression. In human SLP-76 mRNA is abundantly expressed in human spleen, thymus and peripheral blood leucocytes, T-cells and monocytic cell lines, but a low level of expression in B cells.

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