

DESCRIPTION	
Species Reactivity	Human
Specificity	Detects human ZAP70.
Source	Polyclonal Goat IgG
Purification	Antigen Affinity-purified
Immunogen	<i>E. coli</i> -derived recombinant human ZAP70 Trp163-Cys254 Accession # P43403
Formulation	Lyophilized from a 0.2 µm filtered solution in PBS with Trehalose. See Certificate of Analysis for details. *Small pack size (-SP) is supplied either lyophilized or as a 0.2 µm filtered solution in PBS.

APPLICATIONS

Please Note: Optimal dilutions should be determined by each laboratory for each application. General Protocols are available in the Technical Information section on our website.

	Recommended Concentration	Sample
Western Blot	1 µg/mL	See Below
Immunohistochemistry	5-15 µg/mL	See Below
Simple Western	10 µg/mL	See Below

DATA

Western Blot

Detection of Human ZAP70 by Western Blot. Western blot shows lysates of Jurkat human acute T cell leukemia cell line, MOLT-4 human acute lymphoblastic leukemia cell line, and Ramos human Burkitt's lymphoma cell line. PVDF membrane was probed with 1 µg/mL of Human ZAP70 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3709) followed by HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). A specific band was detected for ZAP70 at approximately 70 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunohistochemistry

ZAP70 in Human Lymphoma. ZAP70 was detected in immersion fixed paraffin-embedded sections of human lymphoma using 10 µg/mL Human ZAP70 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3709) overnight at 4 °C. Before incubation with the primary antibody tissue was subjected to heat-induced epitope retrieval using Antigen Retrieval Reagent-Basic (Catalog # CTS013). Tissue was stained with the Anti-Goat HRP-DAB Cell & Tissue Staining Kit (brown; Catalog # CTS008) and counterstained with hematoxylin (blue). Specific labeling was localized to the cytoplasm in epithelial cells. View our protocol for Chromogenic IHC Staining of Paraffin-embedded Tissue Sections.

Simple Western

Detection of Human ZAP70 by Simple Western™. Simple Western lane view shows lysates of Jurkat human acute T cell leukemia cell line, MOLT-4 human acute lymphoblastic leukemia cell line, Ramos human Burkitt's lymphoma cell line, and Raji human Burkitt's lymphoma cell line, loaded at 0.2 mg/mL. A specific band was detected for ZAP70 at approximately 66 and 149 kDa (as indicated) using 10 µg/mL of Goat Anti-Human ZAP70 Antigen Affinity-purified Polyclonal Antibody (Catalog # AF3709) followed by 1:50 dilution of HRP-conjugated Anti-Goat IgG Secondary Antibody (Catalog # HAF109). This experiment was conducted under reducing conditions and using the 12-230 kDa separation system.

PREPARATION AND STORAGE

Reconstitution	Reconstitute at 0.2 mg/mL in sterile PBS.
Shipping	The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below. *Small pack size (-SP) is shipped with polar packs. Upon receipt, store it immediately at -20 to -70 °C
Stability & Storage	Use a manual defrost freezer and avoid repeated freeze-thaw cycles. <ul style="list-style-type: none"> • 12 months from date of receipt, -20 to -70 °C as supplied. • 1 month, 2 to 8 °C under sterile conditions after reconstitution. • 6 months, -20 to -70 °C under sterile conditions after reconstitution.

BACKGROUND

ZAP70 (zeta-chain (TCR) associated protein kinase 70 kDa), expressed primarily in T and NK cells, is a Syk family cytosolic protein tyrosine kinase that consists of two N-terminal SH2 domains and a C-terminal tyrosine kinase domain. Upon T cell receptor activation and phosphorylation of TCR ITAMs by Src family kinases, ZAP70 is recruited to phosphorylated ITAM sequences and subsequently phosphorylated on several tyrosine residues. ZAP70 has been implicated in several immune disorders. An autosomal recessive form of SCID in humans has been attributed to a homozygous mutation in the kinase domain of ZAP70. ZAP70 expression also defines an aggressive subset of CLL.