

DESCRIPTION

Species Reactivity	Human
Specificity	Detects human Phospho-Lck (Y505) in direct ELISA and Western Blot.
Source	Monoclonal Mouse IgG _{2A} Clone # 755216
Purification	Protein A or G purified from hybridoma culture supernatant
Immunogen	Phosphopeptide containing the human Lck (Y505) site. Accession # P06239
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 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	0.5 µg/mL	See Below
Immunocytochemistry	8-25 µg/mL	See Below

DATA

Western Blot

Detection of Human Phospho-Lck (Y505) by Western Blot. Western blot shows lysates of Jurkat human acute T cell leukemia cell line untreated (-) or treated (+) with 1 mM Pervanadate (PV) for 10 minutes or 2mM H₂O₂ for 2 minutes. PVDF membrane was probed with 0.5 µg/mL of Mouse Anti-Human Phospho-Lck (Y505) Monoclonal Antibody (Catalog # MAB7369) followed by HRP-conjugated Anti-Mouse IgG Secondary Antibody (Catalog # HAF018). A specific band was detected for Phospho-Lck (Y505) at approximately 62 kDa (as indicated). This experiment was conducted under reducing conditions and using Immunoblot Buffer Group 1.

Immunocytochemistry

Phospho-Lck (Y505) in Jurkat Human Cell Line. Lck phosphorylated at Y505 was detected in immersion fixed Jurkat human acute T cell leukemia cell line stimulated (upper panel) or not (lower panel) with pervanadate using Mouse Anti-Human Phospho-Lck (Y505) Monoclonal Antibody (Catalog # MAB7369) at 10 µg/mL for 3 hours at room temperature. Cells were stained using the NorthernLights™ 557-conjugated Anti-Mouse IgG Secondary Antibody (red; Catalog # NL007) and counterstained with DAPI (blue). Specific staining was localized to cell surfaces and cytoplasm. View our protocol for [Fluorescent ICC Staining of Non-adherent Cells](#).

PREPARATION AND STORAGE

Reconstitution	Sterile PBS to a final concentration of 0.5 mg/mL.
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

Lck (p56lck) is a 56 kDa cytosolic phosphoprotein within the Src family of non-receptor tyrosine kinases. The 509 amino acid (aa) human Lck contains an SH3 domain (aa 61-121), an SH2 domain (aa 127-224) and a protein kinase domain (aa 251-259). Within aa 2-66, human Lck shares 89% aa sequence identity with mouse and rat Lck. A short (363 aa) isoform contains alternate sequence starting at aa 348, while a 539 aa isoform contains inserted sequence after aa 321. Lck interacts with T cell CD4 and CD8 molecules and plays a pivotal role in regulating T cell activation. It can be overexpressed in cancers and functions as an oncogene.