

Alexa Fluor® 488 anti-Lck Phospho (Tyr394) Antibody

Catalog# / Size	933107 / 25 µg 933108 / 100 µg
Clone	A18002D
Regulatory Status	RUO
Other Names	Lsk, p56lck, LCK Proto-Oncogene, Src Family Tyrosine Kinase, Tyrosine-protein kinase LCK, Leukocyte C-terminal Src kinase, LSK, Lymphocyte cell-specific protein-tyrosine kinase, T cell-specific protein-tyrosine kinase
Isotype	Mouse IgG1, κ
Description	The Src family tyrosine kinase p56 ^{Lck} (Lck) is a non-receptor tyrosine kinase that plays a critical role in T cell selection and maturation within the thymus, and also in the function of mature T cells. Lck, which is constitutively bound to cytosolic domains of CD4 and CD8 surface receptors, plays an essential role in T cell receptor (TCR) signaling. Engagement of the TCR with peptide antigen-loaded MHC complexes results in the recruitment of CD4- and CD8-bound Lck to the TCR/CD3 signaling complex. Lck then transphosphorylates TCR-gamma chains and CD3 subunits, thereby activating the TCR/CD3 signaling pathway and leading to the recruitment and subsequent phosphorylation of Zap70 by Lck. Lck also plays an important role in interleukin-2 signaling that regulates the T cell proliferative response. Phosphorylation of Lck by CSK at tyrosine 505 negatively regulates the kinase, and is proposed to generate a closed, inactive conformation of the protein. Conversely, phosphorylation at tyrosine 394 in the activation loop of Lck greatly stimulates enzymatic activity. In many types of cancer, Lck is a proliferative and anti-apoptotic driver, and has been proposed as a target for therapeutic intervention.

Product Details

Verified Reactivity	Human, Mouse
Antibody Type	Monoclonal
Host Species	Mouse
Immunogen	Synthetic peptide corresponding to human Src phosphorylated at tyrosine 419
Formulation	Phosphate-buffered solution, pH 7.2, containing 0.09% sodium azide
Preparation	The antibody was purified by affinity chromatography and conjugated with Alexa Fluor® 488 under optimal conditions.
Concentration	0.5 mg/mL
Storage & Handling	The antibody solution should be stored undiluted between 2°C and 8°C, and protected from prolonged exposure to light. Do not freeze.
Application	ICFC - Quality tested ICC - Verified
Recommended Usage	Each lot of this antibody is quality control tested by intracellular immunofluorescent staining with flow cytometric analysis . For flow cytometric staining, the suggested use of this reagent is ≤ 0.5 µg per million cells in 100 µL volume. For immunocytochemistry, a concentration range of 1.0 - 10.0 µg/mL is recommended. It is recommended that the reagent be titrated for optimal performance for each application. * Alexa Fluor® 488 has a maximum emission of 519 nm when it is excited at 488 nm. Alexa Fluor® and Pacific Blue™ are trademarks of Life Technologies Corporation. View full statement regarding label licenses
Excitation Laser	Blue Laser (488 nm)

Application Notes	This clone may recognize other Src family members. This clone was developed using a synthetic peptide corresponding to human Src phosphorylated at tyrosine 419, which displays high sequence homology with the region surrounding Lck tyrosine 394. To confirm specificity to Lck Phospho (Tyr394), the cell line J.Cam1.6 was used. This cell line is a clonal derivative of Jurkat cells that harbors a truncated LCK allele with defective protein expression (Straus, et al. 1992. Cell. 70:585).
Additional Product Notes	For use in immunocytochemistry (ICC), it is recommended to fix/permeabilize with either of the following: -Fixation buffer (Cat. No. 420801) with 0.5 % Triton X-100 -Fixation buffer (Cat. No. 420801) with 100% ice-cold methanol -100% ice-cold methanol only
RRID	AB_3717155 (BioLegend Cat. No. 933107) AB_3717155 (BioLegend Cat. No. 933108)

Antigen Details

Structure	Lck is a 509 amino acid protein with a predicted molecular weight of 58 kD
Distribution	Plasma-membrane associated/T cells
Function	Protein Tyrosine Kinase
Interaction	CD4, CD8, CD2, ZAP-70, LAT, CD45, CSK, SHP1, FYN
Cell Type	T cells
Biology Area	Cell Biology, Signal Transduction
Molecular Family	Protein Kinases/Phosphatase
Antigen References	1. Phillipsen L, <i>et al.</i> 2017. <i>Sci Signal.</i> 10:eaaf4736. 2. Moogk D, <i>et al.</i> 2016. <i>J Immunol.</i> 197:644. 3. Klammt C, <i>et al.</i> 2015. <i>Nat Immunol.</i> 16:961. 4. Cancer Genome Atlas Network. 2015. <i>Cell.</i> 161:1681. 5. Casas J, <i>et al.</i> 2014. <i>Nat Commun.</i> 5:5624 6. McNeill L, <i>et al.</i> 2007. <i>Immunity.</i> 27:425. 7. Lefebvre DC, <i>et al.</i> 2003. <i>Biochim Biophys Acta.</i> 1650:40. 8. Kabouridis PS. <i>et al.</i> 2003. <i>Biochem J.</i> 371:907. 9. Abraham N, <i>et al.</i> 1990. <i>Mol. Cell. Biol.</i> 10:5197.

Gene ID [3932](#)

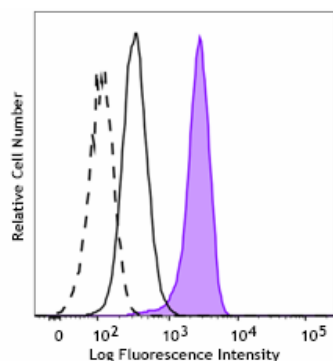
Related Protocols

- [Immunocytochemistry Staining Protocol](#)
- [Intracellular Flow Cytometry Staining Protocol](#)

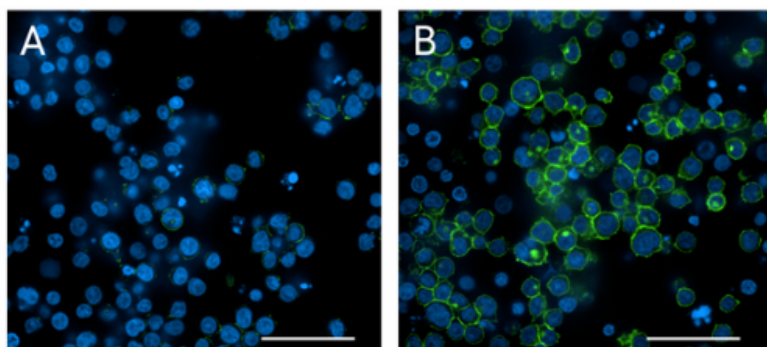
Other Formats

Alexa Fluor® 647 anti-Lck Phospho (Tyr394) , Alexa Fluor® 488 anti-Lck Phospho (Tyr394), Purified anti-Lck Phospho (Tyr394), PE anti-Lck Phospho (Tyr394)

Product Data



Jurkat cells serum-starved overnight and then treated with 0.03% H₂O₂ for 5 minutes to activate Lck (positive cell line, filled histogram) or left untreated (negative cell line, open histogram) were fixed and permeabilized using Cyto-Fast™ Fix/Perm Buffer Set (Cat. No. 426803) and intracellularly stained with Alexa Fluor® 488 Lck Phospho (Tyr394) or Alexa Fluor® 488 Mouse IgG1, κ Isotype Control (open histogram, dashed line, representative histogram for both untreated and treated cells) (Cat. No. 400134).



Jurkat cells were serum starved for 24 hours then treated for 5 minutes with 0.03% H₂O₂ to activate Lck (panel B) or left untreated (panel A). Cells were fixed with 4% PFA Fixation Buffer (Cat. No. 420801) for 10 minutes and permeabilized with 0.5% Triton X-100 for 10 minutes and blocked with 5% FBS for 1 hour at room temperature. Cells were then stained with Alexa Fluor® 488 Lck Phospho (Tyr394) (clone A18002D). Nuclei were counterstained with DAPI (Cat. No. 422801). The images were captured on a Revvity Operetta CLS™ High Content Analysis System with a 63X objective and merged. Scale bar: 50 μm

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