Alexa Fluor® 594 anti-mouse CD4 Antibody

Catalog# / Size	100446 / 100 µg
Clone	GK1.5
Regulatory Status	RUO
Other Names	L3T4, T4
Isotype	Rat IgG2b, κ
Description	CD4 is a 55 kD protein also known as L3T4 or T4. It is a member of the Ig superfamily, primarily expressed on most thymocytes, a subset of T cells, and weakly on macrophages and dendritic cells. It acts as a coreceptor with the TCR during T cell activation and thymic differentiation by binding MHC class II and associating with the protein tyrosin kinase, lck.

Product Details

Verified Reactivity	Mouse
Antibody Type	Monoclonal
Host Species	Rat
Immunogen	Mouse CTL clone V4
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® 594 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	IHC-F - Quality tested FC - Verified SB - Reported in the literature, not verified in house
Recommended Usage	Each lot of this antibody is quality control tested by immunohistochemistry. For immunohistochemistry on frozen tissue sections, a concentration range of 2.5-5 µg/ml is suggested. For flow cytometric staining, the suggested use of this reagent is ≤0.25 µg per million cells in 100 µl volume. It is recommended that the reagent be titrated for optimal performance for each application. * Alexa Fluor® 594 has an excitation maximum of 590 nm, and a maximum emission of 617 nm. Alexa Fluor® and Pacific Blue™ are trademarks of Life Technologies Corporation. View full statement regarding label licenses
Application Notes	Additional reported applications (for the relevant formats) include: blocking of $CD4^+$ T cell activation[1,4,11], thymocyte costimulation[3], *in vitro* and *in vivo* depletion[2,5-8], blocking of egg-sperm cell adhesion[1,4], immunohistochemical staining of acetone-fixed frozen sections[9,10], immunoprecipitation[1,2], and spatial biology (IBEX)[12,13]. The GK1.5 antibody is able to block CD4 mediated cell adhesion and T cell activation. Binding of GK1.5 antibody to CD4 T cells can be blocked by RM4-5 antibody, but not RM4-4 antibody. For *in vivo* studies or highly sensitive assays, we recommend Ultra-LEAF™ purified antibody (Cat. No. 100442) with a lower endotoxin limit than standard LEAF™ purified antibodies (Endotoxin < 0.01 EU/µg).
Additional Product Notes	Iterative Bleaching Extended multi-pleXity (IBEX) is a fluorescent imaging technique capable of highly-multiplexed spatial analysis. The method relies on cyclical bleaching of panels of fluorescent antibodies in order to image and analyze many markers over multiple cycles of staining, imaging, and, bleaching. It is a community-developed open-access method developed by the Center for Advanced Tissue Imaging (CAT-I) in the National Institute of Allergy and Infectious Diseases (NIAID, NIH).

Application References

(PubMed link indicates BioLegend citation)

1. Dialynas DP, *et al.* 1983. *J. Immunol.* 131:2445. (Block, IP)
2. Dialynas DP, *et al.* 1983. *Immunol. Rev.* 74:29. (IP, Deplete)
3. Wu L, *et al.* 1991. *J. Exp. Med.* 174:1617. (Costim)
4. Godfrey DI, *et al.* 1994. *J. Immunol.* 152:4783. (Block)
5. Gavett SH, *et al.* 1994. *Am. J. Respir. Cell. Mol. Biol.* 10:587. (Deplete)
6. Schuyler M, *et al.* 1994. *Am. J. Respir. Crit. Care Med.* 149:1286. (Deplete)
7. Ghobrial RR, *et al.* 1989. *Clin. Immunol. Immunopathol.* 52:486. (Deplete)
8. Israelski DM, *et al.* 1989. *J. Immunol.* 142:954. (Deplete)
9. Zheng B, *et al.* 1996. *J. Exp. Med.* 184:1083. (IHC)
10. Frei K, *et al.* 1997. *J. Exp. Med.* 185:2177. (IHC)
11. Felix NJ, *et al.* 2007. *Nat. Immunol.* 8:388. (Block)
12. Radtke AJ, *et al.* 2020. *Proc Natl Acad Sci U S A.* 117:33455-65. (SB) PubMed
13. Radtke AJ, *et al.* 2022. *Nat Protoc.* 17:378-401. (SB) PubMed

Product Citations

1. Wang D, *et al.* 2018. Immunity. 48:659. PubMed
2. Baptista AP *et al.* 2019. Immunity. 50(5):1188-1201 . PubMed
3. Wang B, *et al.* 2022. Nat Commun. 13:3821. PubMed
4. Ji Y, *et al.* 2017. Mucosal Immunol. 10.1038/mi.2016.119. PubMed
5. Bosnjak B, *et al.* 2019. Front Immunol. 10:840. PubMed
6. Kovacs SB, *et al.* 2021. STAR Protoc. 2:100244. PubMed
7. Tanaka Y, *et al.* 2017. J Immunol. 199:4016. PubMed
8. Kovacs SB, *et al.* 2020. Cell Reports. 32(4):107967. PubMed
9. Liu Q, *et al.* 2021. Adv Mater. 33:e2102852. PubMed
10. Chmielewski M and Abken H 2017. Cell Rep.. 10.1016/j.celrep.2017.11.063. PubMed
11. Wang X, *et al.* 2021. Sci Transl Med. 13:. PubMed

RRID AB_2563182 (BioLegend Cat. No. 100446)

Antigen Details

Structure	Ig superfamily, 55 kD
Distribution	Majority of thymocytes, T cell subset
Function	TCR co-receptor, T cell activation
Ligand/Receptor	MHC class II molecule
Cell Type	Dendritic cells, T cells, Thymocytes, Tregs
Biology Area	Immunology
Molecular Family	CD Molecules

Antigen References

1. Barclay A, *et al.* 1997. The Leukocyte Antigen FactsBook Academic Press.
2. Bierer BE, *et al.* 1989. *Annu. Rev. Immunol.* 7:579.
3. Janeway CA. 1992. *Annu. Rev. Immunol.* 10:645.

Gene ID 12504

Related Protocols

Immunohistochemistry Protocol for Frozen Sections

Cell Surface Flow Cytometry Staining Protocol

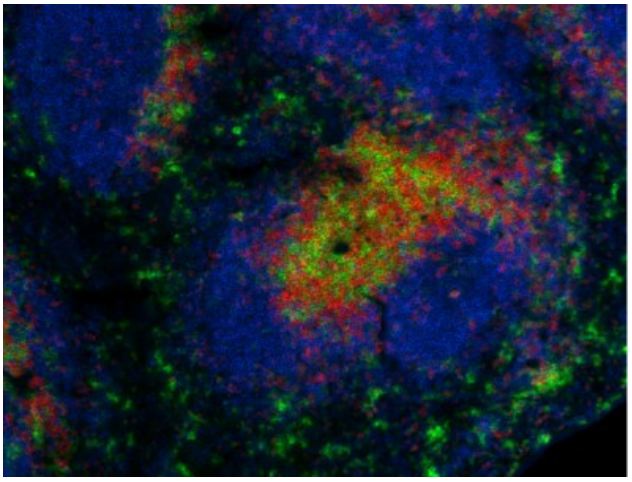
Immunocytochemistry Staining Protocol

Other Formats

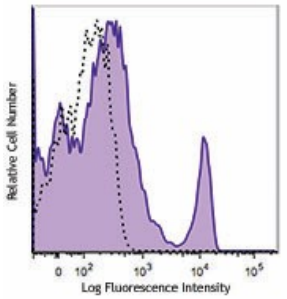
APC anti-mouse CD4, Biotin anti-mouse CD4, FITC anti-mouse CD4, PE anti-mouse CD4, PE/Cyanine5 anti-mouse CD4, Purified anti-mouse CD4, PE/Cyanine7 anti-mouse CD4, APC/Cyanine7 anti-mouse CD4, Alexa Fluor® 647 anti-mouse CD4, Alexa Fluor® 488 anti-mouse CD4, Pacific Blue™ anti-mouse CD4, Alexa Fluor® 700 anti-mouse CD4, PerCP anti-mouse CD4, PerCP/Cyanine5.5 anti-mouse CD4, Brilliant Violet 421™ anti-mouse CD4, Ultra-LEAF™ Purified anti-mouse CD4, Alexa Fluor® 594 anti-mouse CD4, Brilliant Violet 711™ anti-mouse CD4, Brilliant Violet 510™ anti-mouse CD4, Brilliant Violet 605™ anti-mouse CD4,

Brilliant Violet 785™ anti-mouse CD4, PE/Dazzle™ 594 anti-mouse CD4, APC/Fire™ 750 anti-mouse CD4, GoInVivo™ Purified anti-mouse CD4, Brilliant Violet 750™ anti-mouse CD4, Brilliant Violet 650™ anti-mouse CD4, Spark Blue™ 550 anti-mouse CD4, Spark NIR™ 685 anti-mouse CD4, KIRAVIA Blue 520™ anti-mouse CD4, PE/Fire™ 640 anti-mouse CD4, APC/Fire™ 810 anti-mouse CD4, PE/Fire™ 700 anti-mouse CD4, Spark Violet™ 538 anti-mouse CD4, Spark YG™ 593 anti-mouse CD4, Spark Blue™ 574 anti-mouse CD4 Antibody, Spark UV™ 387 anti-mouse CD4

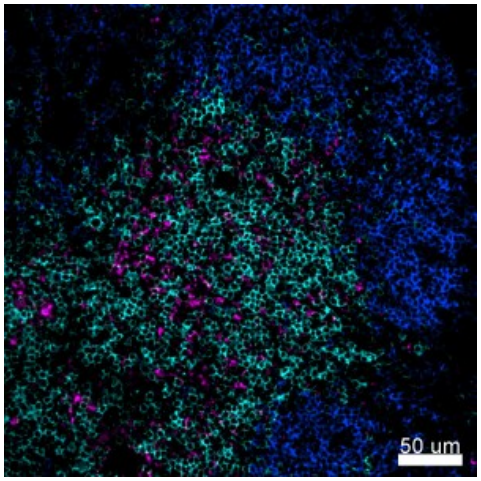
Product Data



C57BL/6 mouse frozen lymph node section was fixed with 4% paraformaldehyde (PFA) for 10 minutes at room temperature and blocked with 5% FBS plus 5% rat serum for 1 hour at room temperature. Then the section was stained with 2.5 µg/ml of CD4 (clone GK1.5) Alexa Fluor® 594 (red), 2.5 µg/ml of CD8 (clone 53-6.7) Alexa Fluor® 647 (green), and 2.5 µg/ml of B220 (clone RA3-6B2) Alexa Fluor® 488 (blue) overnight at 4°C. The image was captured by 10X objective.



C57BL/6 mouse splenocytes were stained with CD4 (clone GK1.5) Alexa Fluor® 594 (filled histogram). The data was acquired by BD LSRFortessa™ cell analyzer equipped with Yellow-Green Laser (561 nm).



Confocal image of C57BL/6 mouse spleen sample acquired using the IBEX method of highly multiplexed antibody-based imaging: CD4 (cyan), CD8 (magenta), and IgD (blue) in Cycle 1. Tissues were prepared using ~1% (vol/vol) formaldehyde and a detergent. Following fixation, samples are immersed in 30% (wt/vol) sucrose for cryoprotection. Images are courtesy of Drs. Andrea J. Radtke and Ronald N. Germain of the Center for Advanced Tissue Imaging (CAT-I) in the National Institute of Allergy and Infectious Diseases (NIAID, NIH).

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