Technical Data Sheet

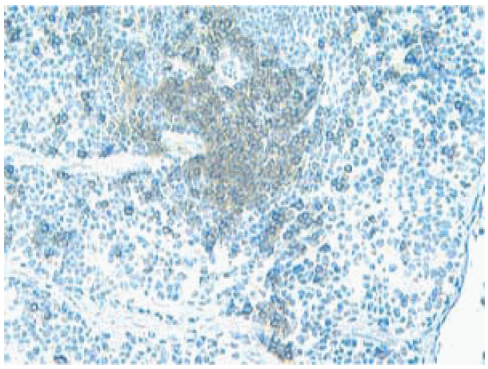
Purified Rat Anti-Mouse CD4

Product Information

Material Number:	**550278**
Alternate Name:	L3T4
Size:	1.0 ml
Concentration:	62.5 µg/ml
Clone:	H129.19
Immunogen:	A.TH mouse CTL clone A15.1.17
Isotype:	Rat (LOU) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

Description

The H129.19 antibody reacts with the CD4 (L3T4) differentiation antigen expressed on thymocytes, a subpopulation of mature T lymphocytes (i.e., MHC class II-restricted T cells, including most T helper cells), and a subset of NK-T cells of all mouse strains tested. CD4 has also been detected on pluirpotent hematopoietic stem cells, bone marrow myeloid precursors, intrathymic lymphoid precursors, and a subset of splenic dendritic cells. CD4 is expressed on the plasma membrane of mouse egg cells and is involved in adhesion of the egg to MHC class II-bearing sperm. CD4 is an antigen coreceptor on the T-cell surface which interacts with MHC class II molecules on antigen-presenting cells. It participates in T-cell activation through its association with the T-cell receptor complex and protein tyrosine lck. H129.19 mAb blocks binding of the anti-mouse CD4 mAbs Gk1.5 (Cat. No. 557307/553729) and RM4-5 (Cat. No. 553046/553047), but not RM4-4 (Cat. No. 553055) antibody. mAb H129.19 inhibits various responses of T helper cells to antigenic or mitogenic stimuli.



Immunohistochemical staining of CD4 + T lymphocytes. *Frozen sections of normal mouse spleen were reacted with H129.19 antibody. CD4+ T lymphocytes can be identified by the intense brown labeling of their cell surface membranes. Amplification 20X.*

Preparation and Storage

Store undiluted at 4°C.

The monoclonal antibody was purified from tissue culture supernatant or ascites by affinity chromatography.

Application Notes

Application

Flow cytometry	Routinely Tested
Immunohistochemistry-frozen	Tested During Development
Immunohistochemistry-zinc-fixed	Tested During Development
Immunoprecipitation	Reported
Blocking	Reported
Depletion	Reported
Immunohistochemistry-formalin (antigen retrieval required)	Not Recommended

Recommended Assay Procedure:

Immunocytochemistry: The H129.19 antibody is recommended to test for immunohistochemical staining of acetone-fixed frozen sections and zinc-fixed paraffin sections. Tissues tested were mouse spleen and thymus. The antibody stains the membranes of thymocytes and a subpopulation of mature T lymphocytes that are MHC class II restricted. The isotype control recommended for use with this antibody is purified rat IgG2a (Cat. No. 559073). For optimal indirect immunohistochemical staining, the H129.19 antibody should be titrated (1:10 to 1:50 dilution) and visualized via a three-step staining procedure in combination with biotinylated polyclonal anti-rat Igs (multiple adsorbed) (Cat. No. 559286) as the secondary

BD Biosciences

bdbiosciences.com

United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	800.979.9408	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

For country contact information, visit **bdbiosciences.com/contact**

antibody and Streptavidin-HRP (Cat. No. 550946) together with the DAB detection system (Cat. No. 550880). More conveniently, the anti-rat Ig HRP detection kit (Cat. No. 551013) that contains the biotinylated secondary antibody, antibody diluent, streptavidin-HRP and DAB substrate can be used for staining. A detailed protocol of the immunohistochemical procedure is available on our website at **www.bdbiosciences.com/support/resources**. The clone H129.19 is not recommended for formalin fixed paraffin embedded sections.

Suggested Companion Products

Catalog Number	Name	Size	Clone
559073	Purified Rat IgG2a κ Isotype Control	0.25 mg	R35-95
559286	Biotin Goat Anti-Rat Ig	0.5 mg	Polyclonal
550880	DAB Substrate Kit	500 tests	(none)
551013	Anti-Rat Ig HRP Detection Kit	200 tests	(none)
550946	Streptavidin HRP	50 ml	(none)

Product Notices

1. Since applications vary, each investigator should titrate the reagent to obtain optimal results.
2. Caution: Sodium azide yields highly toxic hydrazoic acid under acidic conditions. Dilute azide compounds in running water before discarding to avoid accumulation of potentially explosive deposits in plumbing.
3. Source of all serum proteins is from USDA inspected abattoirs located in the United States.
4. An isotype control should be used at the same concentration as the antibody of interest.
5. This antibody has been developed for the immunohistochemistry application. However, a routine immunohistochemistry test is not performed on every lot. Researchers are encouraged to titrate the reagent for optimal performance.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

BD Biosciences Pharmingen. Unpublished results. . (Clone-specific: Immunohistochemistry)

Bierer BE, Sleckman BP, Ratnofsky SE, Burakoff SJ. The biologic roles of CD2, CD4, and CD8 in T-cell activation. *Annu Rev Immunol.* 1989; 7:579-599. (Biology)

Frederickson GG, Basch RS. L3T4 antigen expression by hemopoietic precursor cells. *J Exp Med.* 1989; 169(4):1473-1478. (Biology)

Ghobrial RR, Boublik M, Winn HJ, Auchincloss H Jr. In vivo use of monoclonal antibodies against murine T cell antigens. *Clin Immunol Immunopathol.* 1989; 52(3):486-506. (Clone-specific: Depletion)

Janeway CA Jr. The T cell receptor as a multicomponent signalling machine: CD4/CD8 coreceptors and CD45 in T cell activation. *Annu Rev Immunol.* 1992; 10:645-674. (Biology)

Naquet P, Malissen B, Bekkhoucha F, et al. L3T4 but not LFA-1 participates in antigen presentation by Ak-positive L-cell transformants. *Immunogenetics.* 1985; 22(3):247-256. (Clone-specific: Blocking)

Pierres A, Naquet P, Van Agthoven A, et al. A rat anti-mouse T4 monoclonal antibody (H129.19) inhibits the proliferation of Ia-reactive T cell clones and delineates two phenotypically distinct (T4+, Lyt-2,3-, and T4-, Lyt-2,3+) subsets among anti-Ia cytolytic T cell clones. *J Immunol.* 1984; 132(6):2775-2782. (Immunogen: Blocking, Immunoprecipitation)

Pont S, Regnier-Vigouroux A, Marchetto S, Pierres M. Accessory molecules and T cell activation. II. Antibody binding to L3T4a inhibits Ia-independent mouse T cell proliferation. *Eur J Immunol.* 1987; 17(3):429-432. (Clone-specific: Blocking)

Regnier-Vigouroux A, Blanc D, Pont S, Marchetto S, Pierres M. Accessory molecules and T cell activation. I. Antigen receptor avidity differentially influences T cell sensitivity to inhibition by monoclonal antibodies to LFA-1 and L3T4. *J Immunol.* 1986; 16(11):1385-1390. (Clone-specific: Blocking)

Wineman JP, Gilmore GL, Gritzmacher C, Torbett BE, Muller-Sieburg CE. CD4 is expressed on murine pluripotent hematopoietic stem cells. *Blood.* 1992; 180(7):1717-1724. (Biology)

Wu L, Antica M, Johnson GR, Scollay R, Shortman K. Developmental potential of the earliest precursor cells from the adult mouse thymus. *J Exp Med.* 1991; 174(6):1617-1627. (Biology)

Wu L, Scollay R, Egerton M, Pearse M, Spangrude GJ, Shortman K. CD4 expressed on earliest T-lineage precursor cells in the adult murine thymus. *Nature.* 1991; 349(6304):71-74. (Biology)

BD Biosciences

bdbiosciences.com

United States	Canada	Europe	Japan	Asia Pacific	Latin America/Caribbean
877.232.8995	800.979.9408	32.53.720.550	0120.8555.90	65.6861.0633	55.11.5185.9995

For country contact information, visit **bdbiosciences.com/contact**