

Technical Data Sheet

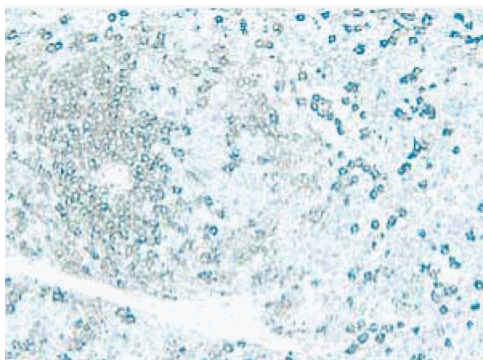
Purified Rat Anti-Mouse CD4

Product Information

Material Number:	550280
Alternate Name:	Cd4; CD4 antigen; L3T4; Ly-4; T-cell surface antigen T4/Leu-3
Size:	1.0 ml
Concentration:	62.5 µg/ml
Clone:	RM4-5
Immunogen:	Mouse Thymocytes (BALB/c)
Isotype:	Rat (DA) IgG2a, κ
Reactivity:	QC Testing: Mouse
Storage Buffer:	Aqueous buffered solution containing BSA, goat serum, and ≤0.09% sodium azide.

Description

The RM4-5 monoclonal antibody specifically binds to the CD4 (L3T4) differentiation antigen expressed on most thymocytes, subpopulations of mature T lymphocytes (i.e., MHC class II-restricted T cells, including most T helper cells and immunosuppressive regulatory T cells), and a subset of NK-T cells. CD4 has also been reported to be detected on pluripotent hematopoietic stem cells, bone marrow myeloid and B-lymphocyte precursors, intrathymic lymphoid precursors, and a subset of splenic dendritic cells. CD4 has been reported to be 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 kinase lck. Purified RM4-5 mAb has been reported to block the binding of FITC-conjugated anti-mouse CD4 clones GK1.5 and H129.19, but not the RM4-4 clone.



Immunohistochemical staining of mouse CD4. Frozen sections of normal mouse spleen were stained with the anti-mouse CD4 antibody clone RM4-5. 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
Immunohistochemistry-formalin (antigen retrieval required)	Not Recommended

Recommended Assay Procedure:

Immunohistochemistry: Clone RM4-5 is recommended to test for immunohistochemical staining of acetone-fixed frozen or zinc-fixed paraffin embedded sections of mouse spleen or thymus. **IHC of formalin-fixed paraffin embedded sections is not recommended.** The isotype control recommended for use with this antibody is purified rat IgG2a (Cat. No. 559073). For optimal indirect immunohistochemical staining, the RM4-5 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 Ig (Cat.No. 559286) as the secondary antibody and streptavidin-HRP (Cat.No. 550946) together with the DAB detection system (Cat.No.

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550880). More conveniently, the anti-rat Ig HRP detection kit (Cat. No. 551013) can be used which contains the biotinylated secondary antibody, the antibody diluent, Streptavidin-HRP and DAB substrates for use in the staining procedure.

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
551013	Anti-Rat Ig HRP Detection Kit	200 tests	(none)
550946	Streptavidin HRP	50 ml	(none)
550880	DAB Substrate Kit	500 tests	(none)
559148	Antibody Diluent for IHC	125 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. 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.
5. An isotype control should be used at the same concentration as the antibody of interest.
6. Please refer to www.bdbiosciences.com/pharmingen/protocols for technical protocols.

References

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