**For Research Use Only.
Not for use in diagnostic procedures.**

MBL
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MONOCLONAL ANTIBODY

Anti-CD4 (Mouse) mAb-FITC

Code No.	Clone	Subclass	Quantity
D341-4	GK1.5	Rat IgG2b κ	1 mL (100 tests)

BACKGROUND: The CD4 antigen is a cell surface glycoprotein that is expressed on subsets of thymocytes and mature T lymphocytes, as well as on monocytes and macrophages. CD4 is an accessory protein for MHC class-II antigen/T-cell receptor interaction and plays an important role in T-helper cell development and activation.
This monoclonal antibody GK1.5 reacts with a non-polymorphic epitope on the mouse CD4 and a useful tool for *in vivo* depletion of $CD4^+$ T cells.

SOURCE: This antibody was purified from mouse ascites fluid. This hybridoma (clone GK1.5) was established by fusion of mouse myeloma cell SP2/0-Ag14 with Lewis rat splenocyte immunized with the cloned cytotoxic T lymphocyte lines V4 and 243/2.5.

FORMULATION: 100 tests in 1 mL volume of PBS containing 1% BSA and 0.09% NaN_3.
*Azide may react with copper or lead in plumbing system to form explosive metal azides. Therefore, always flush plenty of water when disposing materials containing azide into drain.

STORAGE: This antibody solution is stable for one year from the date of purchase when stored at 4°C.

REACTIVITY: This antibody reacts with mouse CD4 antigen on Flow cytometry.

APPLICATION:
Flow cytometry; 10 µL (ready for use)

Detailed procedure is provided in the following **PROTOCOL**.

INTENDED USE:
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SPECIES CROSS REACTIVITY:

Species	Human	Mouse	Rat
Cell	Not tested	splenocyte	Not tested
Reactivity on FCM		+	

REFERENCES:
1) Dialynas, DP., *et al*., *Immunol Rev*. **74**: 29-56 (1983)
2) Dialynas, DP., *et al*., *J. Immunol*. **131**: 2445-2451 (1983)
3) Rice, JC. and Bucy, RP., *J. Immunol*. **154**: 6644-6654 (1995)
4) Buller, RM., *et al*., *Nature* **328**: 77-79 (1987)
5) Ye, X., *et al*., *Gene Ther*. **7**: 1761-1767 (2000)
6) Li, J., *et al*., *Cancer Res*. **63**: 8384-8392 (2003)
7) Janssen, EM., *et al*., *Nature* **434**: 88-93 (2005)
8) Moro, K., *et al*., *Nature* **463**: 540-544 (2010)
9) Murooka, TT., *et al*., *Nature* **490**: 283-287 (2012)

As clone GK1.5 is really famous all over the world, a lot of researches have been reported. These references are a part of such reports.

RELATED PRODUCTS:
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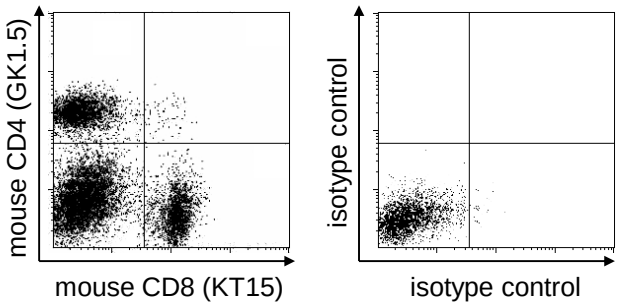
The descriptions of the following protocols are examples. Each user should determine the appropriate condition.



Flow cytometric analysis of mouse CD4 expression (left) and isotypic control (light) on mouse splenocytes. *The staining intensity of D341-4 is shown in the vertical axis with mouse CD8 (clone KT15, MBL PN D271-A64) staining on the horizontal axis.*

PROTOCOL:
Flow cytometric analysis for mouse splenocytes
Single spleen cell suspensions are prepared from the spleens according to standard procedures. We usually use Fisher tubes or equivalents as reaction tubes for all steps described below.
1) Wash the hemolyzed splenocytes 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.05% NaN_3].

2) Resuspend the cells with washing buffer (2 x 10^7 cells/mL).
3) Add 50 μL of the cell suspension into each tube, and centrifuge at 400 x g for 5 minutes at room temperature (20~25°C). Remove supernatant by careful aspiration.
4) Add 10 μL of Clear Back (human Fc receptor blocking reagent, MBL PN MTG-001) to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
5) Add 80 μL of washing buffer containing 10 μL of the anti-CD4 (Mouse) mAb-FITC (clone GK1.5) and 10 μL of the Alexa Fluor® 647 labeled mouse CD8 (clone KT15) to each tube. Mix well and incubate for 30 minutes at 4°C.
6) Add 1 mL of the washing buffer followed by centrifugation at 400 x g for 5 minutes at room temperature. Remove supernatant by careful aspiration.
7) Resuspend the cells with 500 μL of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; mouse splenocytes)