

MONOCLONAL ANTIBODY

Mouse IgG2b (isotype control)-PE

Code No.	Clone	Subclass	Quantity	Concentration
M077-5	3D12	Mouse IgG2b κ	1 mL (50 tests)	10 μ g/mL

SOURCE: This antibody was purified from hybridoma (clone 3D12) supernatant using protein A agarose. This hybridoma was established by fusion of mouse myeloma cell P3U1 with Balb/c mouse lymph nodes immunized with KLH.

FORMULATION: 10 μ g IgG in 1 mL volume of PBS containing 1% BSA and 0.1% ProClin 150.

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

REACTIVITY: No specific binding is detected on human peripheral blood leukocytes.

APPLICATION:

Flow cytometry: 20 μ L (ready for use)

This antibody can be used as a negative isotypic control. The concentration will depend on condition.

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

INTENDED USE:

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

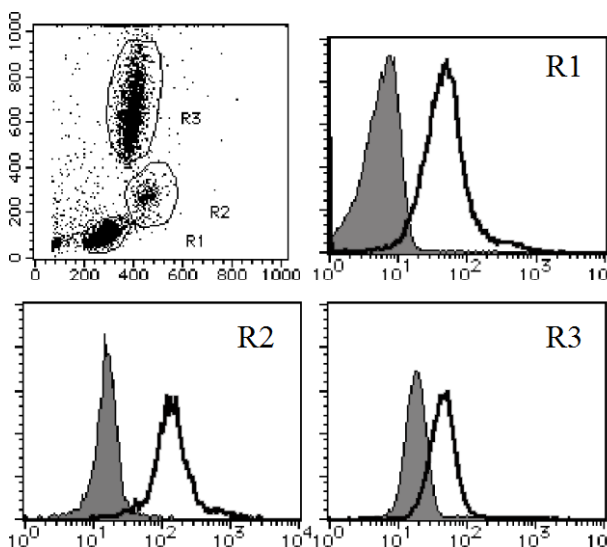
PROTOCOLS:

Flow cytometric analysis for floating cells

We usually use Fisher tubes or equivalents as reaction tubes for all steps described below.

- 1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.09% NaN₃].
*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.
- 2) Resuspend the cells with washing buffer (5x10⁶ cells/mL).
- 3) Add 50 μ L of the cell suspension into each tube, and centrifuge at 500 x g for 1 minute at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 4) Add 20 μ L of Clear Back (human Fc receptor blocking reagent, MBL; code no. MTG-001) to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add the primary antibody at the amount as suggested in the **APPLICATION**. Mix well and incubate for 30 minutes at room temperature.

- 6) Add 1 mL of the washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 7) Resuspend the cells with 500 μ L of the washing buffer and analyze by a flow cytometer.



Flow cytometric analysis of mouse IgG2b reactivity on lymphocyte (R1), monocyte (R2) and granulocyte (R3). Shaded histograms indicate the reaction of M077-5 to the cells. Open histograms indicate the reaction of PE labeled anti-HLA-A24 (K0209-5) to the cells.

Flow cytometric analysis for whole blood cells

We usually use Falcon tubes or equivalents as reaction tubes for all steps described below.

- 1) Add the primary antibody at the amount as suggested in the **APPLICATION** into each tube.
- 2) Add 100 μ L of whole blood into each tube. Mix well and incubate for 30 minutes at room temperature (20~25°C).
- 3) Add 1 mL of the washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.09% NaN₃] followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 4) Lyse with OptiLyse C (for analysis on Beckman Coulter instruments) or OptiLyse B (for analysis on BD instruments), using the procedure recommended in the respective package inserts.
- 5) Add 1 mL of H₂O to each tube and incubate for 10 minutes at room temperature.

- 6) Centrifuge at 500 x g for 1 minute at room temperature.
- 7) Add 1 mL of washing buffer followed by centrifugation at 500 x g for 1 minute at room temperature. Remove supernatant by careful aspiration.
- 8) Resuspend the cells with 500 μ L of the washing buffer and analyze by a flow cytometer.

RELATED PRODUCTS:

Purified antibodies

M075-3	Mouse IgG1 (isotype control) (2E12)
M075-4	Mouse IgG1 (isotype control)-FITC (2E12)
M075-5	Mouse IgG1 (isotype control)-PE (2E12)
M075-6	Mouse IgG1 (isotype control)-Biotin (2E12)
M075-A48	Mouse IgG1 (isotype control)-Alexa Fluor [®] 488 (2E12)
M075-A64	Mouse IgG1 (isotype control)-Alexa Fluor [®] 647 (2E12)
M075-8	Mouse IgG1 (isotype control)-Agarose (2E12)
M076-3	Mouse IgG2a (isotype control) (6H3)
M076-4	Mouse IgG2a (isotype control)-FITC (6H3)
M076-5	Mouse IgG2a (isotype control)-PE (6H3)
M076-6	Mouse IgG2a (isotype control)-Biotin (6H3)
M076-A48	Mouse IgG2a (isotype control)-Alexa Fluor [®] 488 (6H3)
M076-A64	Mouse IgG2a (isotype control)-Alexa Fluor [®] 647 (6H3)
M077-3	Mouse IgG2b (isotype control) (3D12)
M077-4	Mouse IgG2b (isotype control)-FITC (3D12)
M077-5	Mouse IgG2b (isotype control)-PE (3D12)
M077-6	Mouse IgG2b (isotype control)-Biotin (3D12)
M077-A48	Mouse IgG2b (isotype control)-Alexa Fluor [®] 488 (3D12)
M077-A64	Mouse IgG2b (isotype control)-Alexa Fluor [®] 647 (3D12)
M078-3	Mouse IgG3 (isotype control) (6A3)
M078-4	Mouse IgG3 (isotype control)-FITC (6A3)
M078-6	Mouse IgG3 (isotype control)-Biotin (6A3)
M079-3	Mouse IgM (isotype control) (7E10)
M080-3	Rat IgG1 (isotype control) (1H5)
M080-4	Rat IgG1 (isotype control)-FITC (1H5)
M080-5	Rat IgG1 (isotype control)-PE (1H5)
M080-A48	Rat IgG1 (isotype control)-Alexa Fluor [®] 488 (1H5)
M080-A64	Rat IgG1 (isotype control)-Alexa Fluor [®] 647 (1H5)
M081-3	Rat IgG2a (isotype control) (2H3)
M081-4	Rat IgG2a (isotype control)-FITC (2H3)
M081-5	Rat IgG2a (isotype control)-PE (2H3)
M081-A48	Rat IgG2a (isotype control)-Alexa Fluor [®] 488 (2H3)
M081-A64	Rat IgG2a (isotype control)-Alexa Fluor [®] 647 (2H3)
M081-8	Rat IgG2a (isotype control)-Agarose (2H3)
M082-3	Rat IgG2c (isotype control) (6E12)
M082-4	Rat IgG2c (isotype control)-FITC (6E12)
M090-3	Rat IgG2b (isotype control) (3G8)
M090-4	Rat IgG2b (isotype control)-FITC (3G8)
M090-5	Rat IgG2b (isotype control)-PE (3G8)
M090-A48	Rat IgG2b (isotype control)-Alexa Fluor [®] 488 (3G8)
M090-A64	Rat IgG2b (isotype control)-Alexa Fluor [®] 647 (3G8)
PM035	Normal Rabbit IgG (polyclonal)
PM035-8	Normal Rabbit IgG-Agarose (polyclonal)
PM067	Normal Guinea Pig IgG (polyclonal)
M189-3	Syrian Hamster IgG (isotype control)
M199-3	Armenian Hamster IgG (isotype control)
PM084	Normal Chicken IgY (polyclonal)
PM084-4	Normal Chicken IgY-FITC (polyclonal)
PM089	Normal Sheep IgG (polyclonal)
PM094	Normal Goat IgG (polyclonal)

Smart-IP series

3190	Magnetic Rack
M075-11	Mouse IgG1 (isotype control)-Magnetic Beads (2E12)
M076-11	Mouse IgG2a (isotype control)-Magnetic Beads (6H3)
M077-11	Mouse IgG2b (isotype control)-Magnetic Beads (3D12)
M081-11	Rat IgG2a (isotype control)-Magnetic Beads (2H3)
M180-11	Anti-HA-tag mAb-Magnetic Beads (TANA2)
M132-11	Anti-HA-tag mAb-Magnetic Beads (5D8)
M185-11	Anti-DDDDK-tag mAb-Magnetic Beads (FLA-1)
M047-11	Anti-Myc-tag mAb-Magnetic Beads (PL14)
D291-11	Anti-His-tag mAb-Magnetic Beads (OGHis)
D153-11	Anti-GFP mAb-Magnetic Beads (RQ2)
M165-11	Anti-RFP mAb-Magnetic Beads (3G5)
M198-9	Anti-E-tag mAb-Magnetic beads (21D11)
M167-11	Anti-V5-tag mAb-Magnetic Beads (1H6)
D058-9	Anti-Multi Ubiquitin mAb-Magnetic beads (FK2)
M180-10	Anti-HA-tag mAb-Magnetic Agarose (TANA2)
M132-10	Anti-HA-tag mAb-Magnetic Agarose (5D8)
M185-10	Anti-DDDDK-tag mAb-Magnetic Agarose (FLA-1)
M047-10	Anti-Myc-tag mAb-Magnetic Agarose (PL14)
D291-10	Anti-His-tag mAb-Magnetic Agarose (OGHis)
D153-10	Anti-GFP mAb-Magnetic Agarose (RQ2)
M165-10	Anti-RFP mAb-Magnetic Agarose (3G5)
M167-10	Anti-V5-tag mAb-Magnetic Agarose (1H6)
M198-10	Anti-E-tag mAb-Magnetic Agarose (21D11)
M201-10	Anti-Phosphotyrosine mAb-Magnetic Agarose (PT4)

Functional grade antibodies

M075-3M2	Mouse IgG1 (isotype control) (2E12)
M076-3M2	Mouse IgG2a (isotype control) (6H3)
M077-3M2	Mouse IgG2b (isotype control) (3D12)
M078-3M2	Mouse IgG3 (isotype control) (6A3)
M079-3M2	Mouse IgM (isotype control) (7E10)
M080-3M2	Rat IgG1 (isotype control) (1H5)
M081-3M2	Rat IgG2a (isotype control) (2H3)
M090-3M2	Rat IgG2b (isotype control) (3G8)