

MONOCLONAL ANTIBODY

Mouse IgG2b (isotype control)-Alexa Fluor[®] 488

Code No.	Clone	Subclass	Quantity	Concentration
M077-A48	3D12	Mouse IgG2b κ	100 μ L	1 mg/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: 100 μ g IgG in 100 μ L 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 lymphocyte, monocyte and granulocyte.

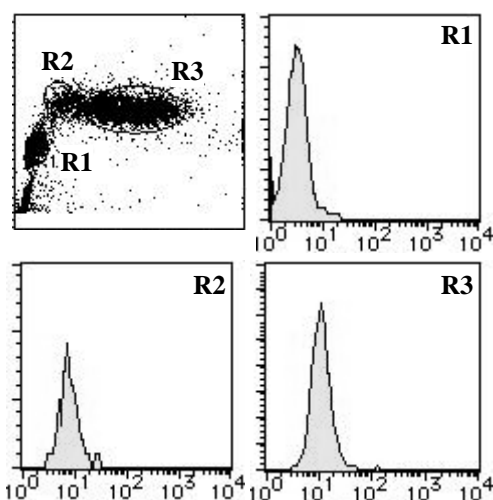
APPLICATION:

Flow cytometry: 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.



Flow cytometric analysis of mouse IgG2b reactivity on lymphocyte (R1), monocyte (R2) and granulocyte (R3).

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 (5 x 10⁶ 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 isotype control antibody at the concentrations comparable to those of the specific antibody of interest. 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 for whole blood cells

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

- 1) Add the isotype control antibody into each tube at the concentrations comparable to those of the specific antibody of interest.
- 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:

[Functional grade antibodies]

M075-3M2 Mouse IgG1 (isotype control) FG (2E12)
M076-3M2 Mouse IgG2a (isotype control) FG (6H3)
M077-3M2 Mouse IgG2b (isotype control) FG (3D12)
M080-3M2 Rat IgG1 (isotype control) FG (1H5)
M081-3M2 Rat IgG2a (isotype control) FG (2H3)
M090-3M2 Rat IgG2b (isotype control) FG (3G8)

[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)
M075-7 Mouse IgG1 (isotype control)-HRP-DirecT (2E12)
M075-8 Mouse IgG1 (isotype control)-Agarose (2E12)
M075-11 Mouse IgG1 (isotype control)-Magnetic Beads (2E12)
M075-12 Mouse IgG1 (isotype control)-ALP (2E12)
M075-A48 Mouse IgG1 (isotype control)-Alexa Fluor[®] 488 (2E12)
M075-A64 Mouse IgG1 (isotype control)-Alexa Fluor[®] 647 (2E12)
M076-3 Mouse IgG2a (isotype control) (6H3)
M076-4 Mouse IgG2a (isotype control)-FITC (6H3)
M076-5 Mouse IgG2a (isotype control)-PE (6H3)
M076-5 Mouse IgG2a (isotype control)-PE (6H3)
M076-11 Mouse IgG2a (isotype control)-Magnetic Beads (6H3)
M076-12 Mouse IgG2a (isotype control)-ALP (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)
M077-11 Mouse IgG2b (isotype control)-Magnetic Beads (3D12)
M077-12 Mouse IgG2b isotype control-ALP (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)
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)
M081-3 Rat IgG2a (isotype control) (2H3)
M081-4 Rat IgG2a (isotype control)-FITC (2H3)
M081-5 Rat IgG2a (isotype control)-PE (2H3)
M081-8 Rat IgG2a (isotype control)-Agarose (2H3)
M081-11 Rat IgG2a (isotype control)-Magnetic Beads (2H3)
M081-A48 Rat IgG2a (isotype control)-Alexa Fluor[®] 488 (2H3)
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)
M082-3 Rat IgG2c (isotype control) (6E12)

M082-4 Rat IgG2c (isotype control)-FITC (6E12)
M189-3 Syrian Hamster IgG (isotype control) (ttko)
M199-3 Armenian Hamster IgG (isotype control) (ttko2)
M194-3 Human IgG1 isotype control chimeric mAb (2E12G1-2)
M195-3 Human IgG2 isotype control chimeric mAb (2E12G2-18)
PM053 Normal Rabbit IgG (polyclonal)
PM035-8 Normal Rabbit IgG-Agarose (polyclonal)
PM067 Normal Guinea Pig IgG (polyclonal)
PM084 Normal Chicken IgY (polyclonal)
PM084-4 Normal Chicken IgY-FITC (polyclonal)
PM084-5 Normal Chicken IgY-PE (polyclonal)
PM089 Normal Sheep IgG (polyclonal)
PM094 Normal Goat IgG (polyclonal)

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LABEL LICENSES:

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