

MONOCLONAL ANTIBODY

Alexa Fluor[®] 488 labeled Mouse IgG2a isotype control

Code No.	Clone	Subclass	Quantity	Concentration
M076-A48	6H3	Mouse IgG2a κ	100 μ L	1 mg/mL

SOURCE: This antibody was purified from hybridoma (clone 6H3) 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 0.09% 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.

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.

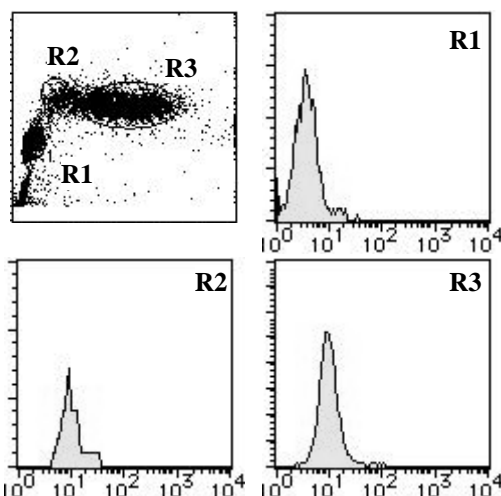
APPLICATION:

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 IgG2a 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 0.1% NaN₃].
- 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 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 0.1% 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 antibody]

- 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 antibody]

- M075-3 Mouse IgG1 isotype control (2E12)
- M075-4 Mouse IgG1 isotype control-FITC (2E12)
- M075-5 Mouse IgG1 isotype control-PE (2E12)
- M075-8 Mouse IgG1 isotype control-Agarose (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-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-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-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)
- PM035-8 Normal Rabbit IgG-Agarose (polyclonal)

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