

MONOCLONAL ANTIBODY

PE labeled Rat IgG2a Isotype control

Code No.	Clone	Subclass	Quantity	Concentration
M081-5	2H3	Rat IgG2a	1 mL (50 tests)	10 µg/mL

SOURCE: This antibody was purified from hybridoma (clone 2H3) supernatant using protein G agarose. This hybridoma was established by fusion of mouse myeloma cell P3U1 with rat lymph nodes immunized with KLH.

FORMULATION: 50 tests in 1 mL 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 detected on mouse splenocyte.

APPLICATION:

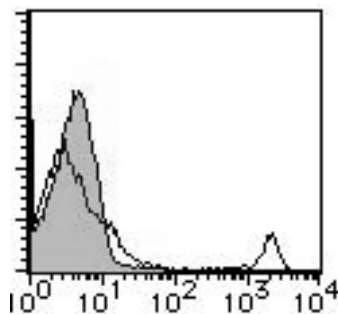
Flow cytometry: 20 µL (ready for use)

This antibody can be used as negative isotypic control. The concentration of antibody 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 PE labeled rat IgG2a on mouse splenocyte. Shaded histogram indicates the reaction of M081-5 to the cells. Open histogram indicates the reaction of mouse PE labeled CD8 to the cells.

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 µL 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 10 µL of normal goat serum containing 1 mg/mL normal human IgG and 0.1% NaN₃ to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
- 5) Add 20 µL of the PE labeled Rat IgG2a Isotype control. Mix well and incubate for 20 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 µL of the washing buffer and analyze by a flow cytometer.

Flow cytometric analysis for whole blood cells

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

- 1) Add 20 µL of PE labeled Rat IgG2a Isotype control into each tube.
- 2) Add 50 µL of whole blood into each tube. Mix well, and incubate for 30 minutes at room temperature (20~25°C).
- 3) Add 1 mL of 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. Remove supernatant by careful aspiration.
- 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 µL of the washing buffer and analyze by a flow cytometer.

RELATED PRODUCTS:

- M075-3 Mouse IgG1 Isotype control (2E12)
- M075-4 FITC labeled Mouse IgG1 Isotype control (2E12)
- M075-5 PE labeled Mouse IgG1 isotype control (2E12)
- M075-8 Agarose conjugated Mouse IgG1 Isotype control (2E12)
- M076-3 Mouse IgG2a Isotype control (6H3)
- M076-4 FITC labeled Mouse IgG2a Isotype control (6H3)
- M076-5 PE labeled Mouse IgG2a isotype control (6H3)
- M077-3 Mouse IgG2b Isotype control (3D12)
- M077-4 FITC labeled Mouse IgG2b Isotype control (3D12)
- M077-5 PE labeled Mouse IgG2b isotype control (3D12)
- M078-3 Mouse IgG3 Isotype control (6A3)
- M078-4 FITC labeled Mouse IgG3 Isotype control (6A3)
- M079-3 Mouse IgM Isotype control (7E10)
- M080-3 Rat IgG1 Isotype control (1H5)
- M080-4 FITC labeled Rat IgG1 Isotype control (1H5)
- M080-5 PE labeled Rat IgG1 Isotype control (1H5)
- M081-3 Rat IgG2a Isotype control (2H3)
- M081-4 FITC labeled Rat IgG2a Isotype control (2H3)
- M081-5 PE labeled Rat IgG2a Isotype control (2H3)
- M081-8 Agarose conjugated Rat IgG2a Isotype control (2H3)
- M090-3 Rat IgG2b Isotype control (3G8)
- M090-4 FITC labeled Rat IgG2b Isotype control (3G8)
- M082-3 Rat IgG2c Isotype control (6E12)
- M082-4 FITC labeled Rat IgG2c Isotype control (6E12)
- PM035-8 Agarose conjugated Normal Rabbit IgG (polyclonal)