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

MONOCLONAL ANTIBODY

FITC labeled Rat IgG1 Isotype control

Code No.	Clone	Subclass	Quantity	Concentration
M080-4	1H5	Rat IgG1	1 mL	50 μg/mL

SOURCE: This antibody was purified from hybridoma (clone 1H5) 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 μg IgG 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: No specific binding detected on human peripheral blood leukocytes.

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:
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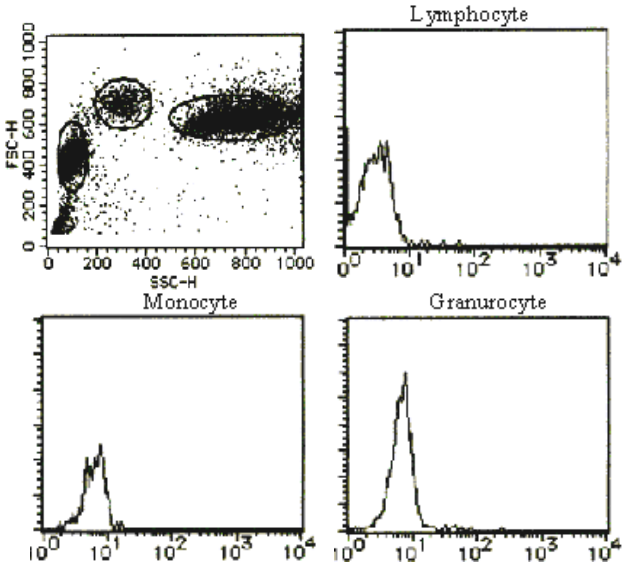
RELATED PRODUCTS:

M075-3	Mouse IgG1 Isotype control (2E12)
M075-4	FITC 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)
M077-3	Mouse IgG2b Isotype control (3D12)
M077-4	FITC 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)
M081-3	Rat IgG2a Isotype control (2H3)
M081-4	FITC 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)



Flow cytometric analysis of FITC labeled Rat IgG1 Isotype control (M080-4) reactivity on human peripheral blood leukocytes.

PROTOCOLS:

Flow cytometric analysis for floating cells

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

1) Wash the cells 3 times with washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN_3].
2) Resuspend the cells with washing buffer ($5x10^6$ 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% NaN_3 to the cell pellet after tapping. Mix well and incubate for 5 minutes at room temperature.
5) Add 40 μL of the FITC labeled Rat IgG1 Isotype control (10-20 μg/mL) diluted with the washing buffer. 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 μL of the washing buffer and analyze by a flow cytometer.

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Flow cytometric analysis for whole blood cells

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

1) Add 50 μL of FITC labeled Rat IgG1 Isotype control (10-20 μg/mL) diluted with the washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.1% NaN_3] 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 ^oC).
3) Add 1 mL of washing buffer 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 H_2O 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.