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



Anti-FcεR1γ (FcRγ) (Mouse) mAb -Alexa Fluor[®] 647

CODE No.

M191-A64

CLONALITY	Monoclonal
CLONE	1D6
ISOTYPE	Mouse IgG1 κ
QUANTITY	100 µL, 1 mg/mL

SOURCEPurified IgG from hybridoma supernatantFORMULATIONPBS 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.

APPLICATIONS-CONFIRMED

Immunocytochemistry	1 μg/mL
Flow cytometry	1 μg/mL

SPECIES CROSS REACTIVITY

Species	Human	Mouse	Rat	Hamster
Cell	Not tested	RAW264, Mouse peritoneal macrophage	Not tested	Not Tested
Reactivity		+		

Entrez Gene ID 14127 (Mouse)

REFERENCES 1) Yamasaki, S., et al., Nat. Immunol. 9, 1179-1188 (2008)

 2) Cao, L., et al., J.Immunol. 179, 5864-5876 (2007)

 3) Sato, K., et al., J. Biol. Chem. 281, 38854-38866 (2006)

 4) Ra, C., et al., J. Biol. Chem. 264, 15323-15327 (1989)

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LABEL LICENSES:

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The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

Immunocytochemistry

- 1) Spread the cells on a glass slide, then incubate in a CO₂ incubator for one night.
- 2) Remove the culture supernatant by careful aspiration.
- 3) Fix the cells by immersing the slide in 4% paraformaldehyde (PFA)/PBS for 10 min. at room temperature (20~25°C).
- 4) Prepare a wash container such as a 500 mL beaker with a magnetic stirrer. Then wash the fixed cells on the glass slide by soaking the slide with a plenty of PBS in the wash container for 5 min. Take care not to touch the cells. Repeat another wash once more.
- 5) Immerse the slide in 0.2% Triton X-100/PBS for 10 min. at room temperature.
- 6) Wash the slide twice in PBS for 5 min. each.
- 7) Add Clear Back (human Fc receptor blocking reagent, MBL; code no. MTG-001) to the cell and incubate for 5 min. at room temperature.
- 8) Add the primary antibody diluted with 2% fetal calf serum (FCS)/PBS as suggested in the **APPLICATIONS** onto the cells and incubate for 1 hr. at room temperature. (Optimization of antibody concentration or incubation condition is recommended if necessary.)
- 9) Wash the slide twice in PBS for 5 min. each.
- 10) Wipe excess liquid from slide but take care not to touch the cells. Never leave the cells to dry.
- 11) Counter stain with DAPI for 5 min. at room temperature.
- 12) Wash the slide twice in PBS for 5 min. each.
- 13) Promptly add mounting medium onto the slide, then put a cover slip on it.

(Positive control for Immunocytochemistry; RAW264)



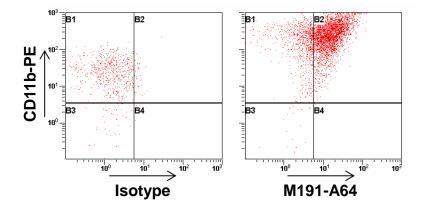
Immunocytochemical detection of FcεR1γ (Mouse) in RAW264 Magenta: M191-A64 Cyan: DAPI

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Flow cytometric analysis

- 1) Wash the cells (5 x 10⁵ cells/sample) 3 times with 1 mL of washing buffer (PBS containing 2% fetal calf serum (FCS)).
- 2) Add 4% paraformaldehyde (PFA)/PBS to the cell pellet after tapping. Mix well, then fix the cells for 10 min. at room temperature.
- 3) Wash the cells twice with 1 mL of washing buffer.
- 4) Add 0.2% Triton X-100 in PBS to the cell pellet after tapping. Mix well, then permeabilize the cells for 10 min. at room temperature.
- 5) Wash the cells twice with 1 mL of washing buffer.
- 6) Add 20 μL 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 min. at room temperature.
- 7) Add 40 µL of the primary antibody at the concentration as suggested in the APPLICATIONS diluted in the washing buffer Mix well and incubate for 30 min. at room temperature.
- 8) Wash the cells 1 time with 1 mL of washing buffer.
- Add 40 μL of 1:200 anti-CD11b (Mouse)-PE (Beckman Coulter; code no. 732048) diluted in the washing buffer. Mix well
 and incubate for 30 min. at room temperature.
- 10) Wash the cells 1 time with 1 mL of washing buffer.
- 11) Resuspend the cells with 500 μ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; Mouse peritoneal macrophage)



Flow cytometric detection of $Fc \in R1\gamma$ (Mouse) in

mouse peritoneal macrophage

Left: isotype control (M075-A64) Right: M191-A64