

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

# Anti-FcεR1γ (FcRγ) (Mouse) mAb -Alexa Fluor® 488

|                    |   |
|--------------------|---|
| <b>CODE No.</b>    | M191-A48  |
| <b>CLONALITY</b>   | Monoclonal  |
| <b>CLONE</b>       | 1D6   |
| <b>ISOTYPE</b>     | Mouse IgG1 κ  |
| <b>QUANTITY</b>    | 100 μL, 1 mg/mL   |
| <b>SOURCE</b>      | Purified IgG from hybridoma supernatant   |
| <b>FORMULATION</b> | PBS containing 1% BSA and 0.1% ProClin 150  |
| <b>STORAGE</b>     | This antibody solution is stable for one year from the date of purchase when stored at 4°C. |

## APPLICATIONS-CONFIRMED

|                            |         |
|----------------------------|---------|
| <u>Immunocytochemistry</u> | 1 μg/mL |
| <u>Flow cytometry</u>      | 1 μg/mL |

## SPECIES CROSS REACTIVITY

| Species    | Human      | Mouse                               | Rat        | Hamster    |
|------------|------------|-------------------------------------|------------|------------|
| Cells      | 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)

For more information, please visit our website at <https://ruo.mbl.co.jp/>.

## LABEL LICENSES:

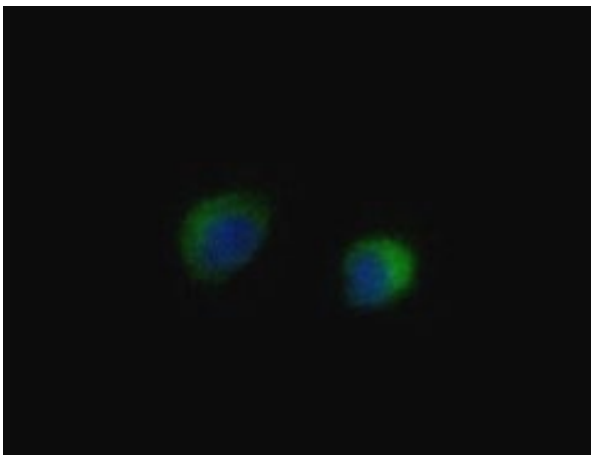
This product is provided under an agreement between LIFE TECHNOLOGIES Corporation, and Medical & Biological Laboratories Co., LTD. Alexa Fluor® is a registered trademark of Molecular Probes, Inc..

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 CO<sub>2</sub> incubator overnight.
- 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 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)



### ***Immunocytochemistry detection of mouse FcεR1γ***

Cell: RAW264

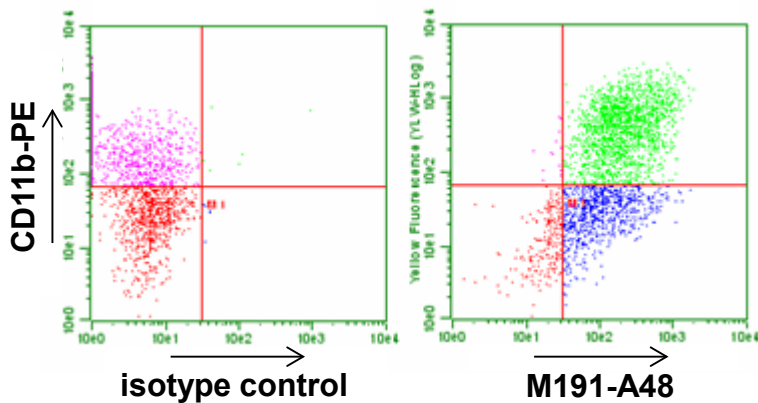
Green: Anti-FcεR1γ (FcRγ) (Mouse) mAb (MBL, code no. M191-A48)

Blue: DAPI

**Flow cytometric analysis**

- 1) Wash the cells ( $5 \times 10^5$  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  $\mu$ 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 min. at room temperature.
- 7) Add 40  $\mu$ L 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 once with 1 mL of washing buffer.
- 9) Add PE-labeled anti-mouse CD11b antibody diluted in the washing buffer. Mix well and incubate for 30 min. at room temperature.
- 10) Wash the cells once with 1 mL of washing buffer.
- 11) Resuspend the cells with 500  $\mu$ L of the washing buffer and analyze by a flow cytometer.

(Positive control for Flow cytometry; Mouse peritoneal macrophage)



***Flow cytometric detection of mouse Fc $\epsilon$ R1 $\gamma$***

Cells: Mouse peritonea macrophage

Left: Mouse IgG1 (isotype control) (MBL, code no. M075-A48)

Right: Anti- Fc $\epsilon$ R1 $\gamma$ (Fc $\epsilon$ R $\gamma$ ) (Mouse) mAb (MBL, code no. M191-A48)