

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

# Anti-CD8a (Human) mAb-FITC

**CODE No.** K0226-4

**CLONALITY** Monoclonal  
**CLONE** Hit8a  
**ISOTYPE** Mouse IgG1  $\kappa$   
**QUANTITY** 1 mL

**FORMULATION** 100 tests in 1 mL volume of PBS containing 1% BSA and 0.1% ProClin 150.  
**STORAGE** This antibody solution is stable for one year from the date of purchase when stored at 4°C.

## APPLICATION-CONFIRMED

Flow cytometry 10  $\mu$ L (ready for use)

## SPECIES CROSS REACTIVITY on FCM

Species	Human		Mouse	Rat	Others
Samples	Sup-T1, PBMC	Jurkat	Not tested	Not tested	Not tested
Reactivity	+	-			

**Entrez Gene ID** 925 (Human)

Please visit our web site <https://ruo.mbl.co.jp/>.

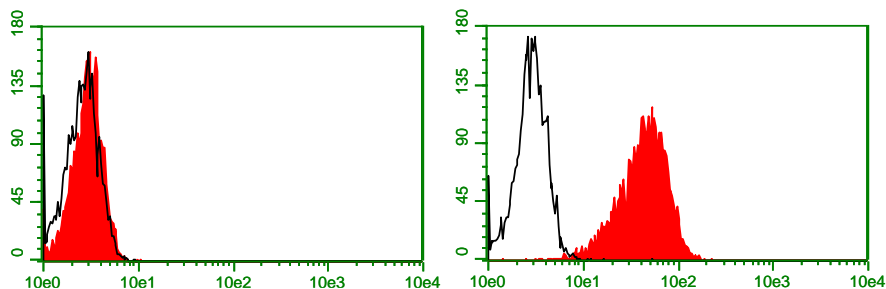
The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

### **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 1 mL of washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.05% NaN<sub>3</sub>].  
\*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.
- 2) Resuspend the cells with washing buffer (3 x 10<sup>6</sup> cells/mL).
- 3) Add 70 µL of the cell suspension into each tube, and centrifuge at 400 x g for 3 minutes at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 4) 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 minutes at room temperature.
- 5) Add 10 µL of Anti-CD8a (Human) mAb-FITC as suggested in the **APPLICATION**. Mix well and incubate for 30 minutes at room temperature.
- 6) Add 1 mL of the washing buffer followed by centrifugation at 400 x g for 3 minutes at room temperature. Remove supernatant by careful aspiration.
- 7) Resuspend the cells with 500 µL of washing buffer containing 1% 7-AAD, and analyze by a flow cytometer.

(Positive control for Flow cytometry; Sup-T1)



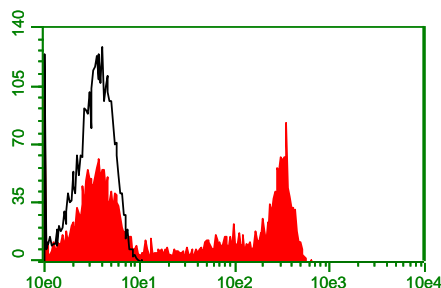
### ***Flow cytometric detection of human CD8a in floating cells***

Cell  
Left: Jurkat  
Right: Sup-T1  
Antibody  
Open: isotype control (M075-4)  
Closed: K0226-4

### **Flow cytometric analysis for floating cells**

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

- 1) Prepare peripheral blood mononuclear cells (PBMC) according to established procedures.
- 2) Wash the cells 3 times with 1 mL of washing buffer [PBS containing 2% fetal calf serum (FCS) and 0.05% NaN<sub>3</sub>].  
\*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.
- 3) Resuspend the cells with washing buffer (3 x 10<sup>6</sup> cells/mL).
- 4) Add 70 µL of the cell suspension into each tube, and centrifuge at 400 x g for 3 minutes at room temperature (20~25°C). Remove supernatant by careful aspiration.
- 5) 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 minutes at room temperature.
- 6) Add 10 µL of Anti-CD8a (Human) mAb-FITC as suggested in the **APPLICATION**. Mix well and incubate for 30 minutes at room temperature.
- 7) Add 1 mL of the washing buffer followed by centrifugation at 400 x g for 3 minutes at room temperature. Remove supernatant by careful aspiration.
- 8) Resuspend the cells with 500 µL of washing buffer containing 1% 7-AAD, and analyze by a flow cytometer.



### ***Flow cytometric detection of human CD8a in PBMC***

Open: isotype control (M075-4)  
Closed: K0226-4