

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

Anti-PE mAb

CODE No.	M240-3
CLONALITY	Monoclonal
CLONE	C27-6
ISOTYPE	Mouse IgG2a κ
QUANTITY	100 μ L, 500 μ g/mL
SOURCE	Purified IgG from hybridoma supernatant
IMMUNOGEN	R-Phycoerythrin (PE)
FORMULATION	PBS containing 50% Glycerol (pH 7.2). No preservative is contained.
STORAGE	This antibody solution is stable for one year from the date of purchase when stored at -20°C.

APPLICATION-CONFIRMED

MHC Tetramer staining 1:50

REFERENCE 1) Tungatt, K., *et al.*, *J. Immunol.* **194**, 463-474 (2015)

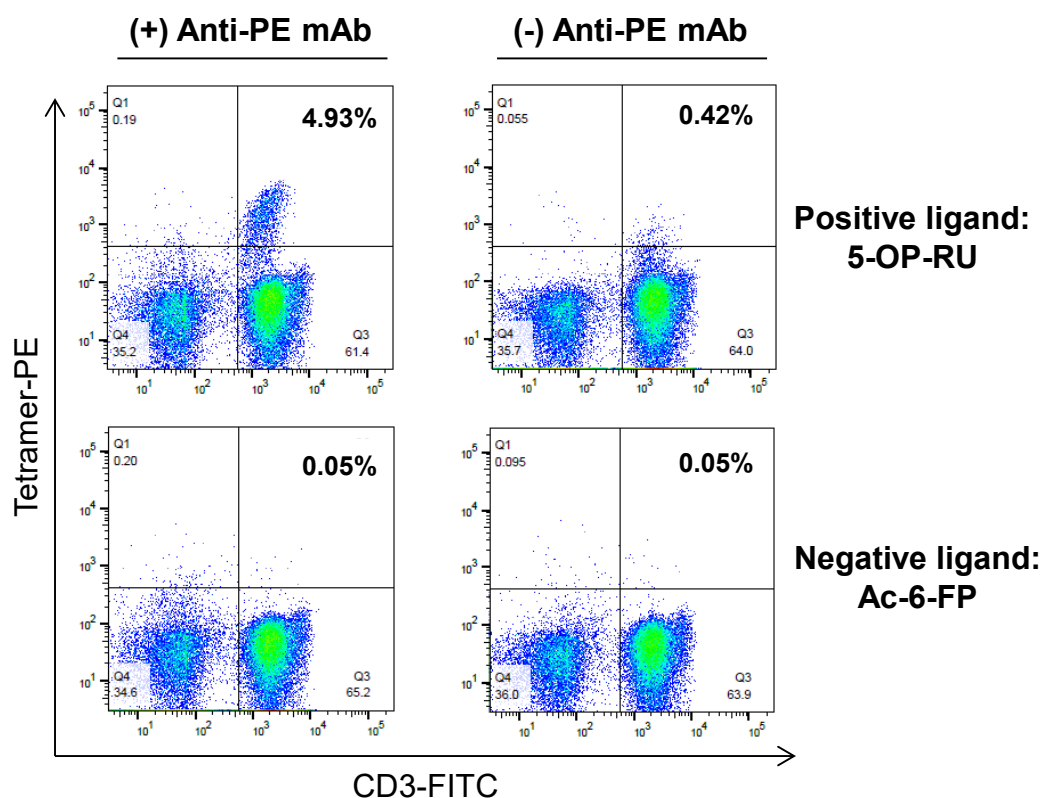
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The descriptions of the following protocols are examples. Each user should determine the appropriate condition.

MHC Tetramer staining

*Before staining, please make sure to check the detailed information of each MHC Tetramer.

- 1) Prepare a single cell suspension from anticoagulated human peripheral blood according to the standard procedure. For staining, suspend the cells in FCM buffer (2% FCS/0.05% NaN₃/PBS) at a concentration up to 2-10 x 10⁵ cells/tube.
 *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) Add 5 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.
- 3) Add 10 µL of T-Select Human MR1 Tetramer v2-PE (MBL, code no. TS-HMRV2-1) loaded with the MR1 ligand and dilute to 50 µL with FCM buffer. Mix well and incubate the cells in the dark for 30-60 minutes at 2-8°C.
- 4) Add 500 µL of FCM buffer and centrifuge at 400 × g for 3 minutes. Aspirate the supernatant.
- 5) Add 1 µL of Anti-PE mAb (MBL, code no. M240-3) as an enhancer and dilute to 50 µL with FCM buffer. Mix well and incubate the cells in the dark for 20 minutes at 2-8°C.
- 6) Wash the cells as step 4).
- 7) Add 5 µL of Anti-CD3 antibody and dilute to 50 µL with FCM buffer. Mix well and incubate the cells in the dark for 30 minutes at 2-8°C.
- 8) Wash the cells twice as step 4).
- 9) Suspend the pellet in 500 µL of FCM buffer containing 7-AAD. Analyze it immediately or suspend it in 0.5% paraformaldehyde/PBS and store the sample in a dark room at 2-8°C. Be sure to analyze it within 24 hours.



Flow cytometric analysis of human PBMCs stained with Human MR1 Tetramer

The data shows the staining of human PBMCs using ligand (5-OP-RU or Ac-6-FP) loading T-Select Human MR1 Tetramer v2-PE (MBL, code no. TS-HMRV2-1) with or without Anti-PE antibody (MBL, code no. M240-3) to enhance staining intensity. Numbers in the upper right quadrants represent the percentages of T-select Human MR1 Tetramer⁺ cells relative to the total CD3⁺ cells.