



# Anti-Ter-119 antibodies mouse

Anti-Ter-119-FITC	130-091-786
Anti-Ter-119-PE	130-091-783
Anti-Ter-119-APC	130-091-788

## Contents

1. Description
  - 1.1 Background information
  - 1.2 Applications
  - 1.3 Recommended antibody dilution
  - 1.4 Reagent requirements
2. General protocol for immunofluorescent staining
3. Examples of immunofluorescent staining with Anti-Ter-119 antibodies
4. References

## 1. Description

<b>Components</b>	1 mL Anti-Ter-119 antibodies, mouse: monoclonal Anti-Ter-119 antibodies conjugated to fluorescein isothiocyanate (FITC), R-phycoerythrin (PE), or allophycocyanin (APC).
<b>Clone</b>	Ter-119 (isotype: rat IgG2b).
<b>Capacity</b>	100 tests or up to $10^9$ total cells.
<b>Product format</b>	Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide.
<b>Storage</b>	Store protected from light at 2–8 °C. Do not freeze. The expiration date is indicated on the vial label.

### 1.1 Background information

The Ter-119 antigen is expressed on mature erythrocytes and erythroid precursor cells in adult blood, lymphoid tissue and bone marrow, and in the embryonic yolk sac and fetal liver.<sup>1-3</sup> The Anti-Ter-119 antibody does not react with cells showing typical erythroid blast-forming unit (BFU-E) and erythroid colony-forming unit (CFU-E) activity.<sup>1</sup> In adult mice, Ter-119 reacts with 20–25% of bone marrow cells and 2–3% of spleen cells but not with thymocytes or lymph node cells.<sup>1</sup>

### 1.2 Applications

- Identification and enumeration of Anti-Ter-119<sup>+</sup> cells by flow cytometry or fluorescence microscopy.
- Evaluation of MACS® Separations by flow cytometry or fluorescence microscopy, for example:
  - Positive selection or depletion of mouse erythroid cells by using Anti-Ter-119 MicroBeads, mouse (# 130-049-901);
  - Positive selection or depletion of mouse Ter-119<sup>+</sup> early erythroid cells from hematopoietic precursor cells by using the Lineage Cell Depletion Kit, mouse (# 130-090-858) in combination with Anti-Sca-1 MicroBead Kit (FITC), mouse (# 130-092-529) or CD117 MicroBeads, mouse (# 130-091-224).

### 1.3 Recommended antibody dilution

For antibody labeling of mouse cells.

Anti-Ter-119 conjugate	FITC	PE	APC
<b>Flow cytometry<sup>a</sup></b>			
- In general	1:11	1:11	1:11
- Anti-Ter-119 MicroBead-labeled cells	1:11	1:11	1:11

a) Given antibody dilutions are for a cell concentration of up to  $10^7$  cells/100  $\mu$ L of buffer.

### 1.4 Reagent requirements

- Buffer: Prepare a solution containing phosphate-buffered saline (PBS) pH 7.2, 0.5% bovine serum albumin (BSA), and 2 mM EDTA by diluting MACS BSA Stock Solution (# 130-091-376) 1:20 with autoMACS™ Rinsing Solution (# 130-091-222). Keep buffer cold (2–8 °C).
  - ▲ Note: EDTA can be replaced by other supplements such as anticoagulant citrate dextrose formula-A (ACD-A) or citrate phosphate dextrose (CPD). BSA can be replaced by other proteins such as mouseserum albumin, mouse serum, or fetal bovine serum. Buffers or media containing Ca<sup>2+</sup> or Mg<sup>2+</sup> are not recommended for use.
- (Optional) Propidium iodide (PI) or 7-AAD for flow cytometric exclusion of dead cells without fixation.
- (Optional) Fixation and Dead Cell Discrimination Kit (# 130-091-163) for cell fixation and flow cytometric exclusion of dead cells.

## 2. General protocol for immunofluorescent staining

▲ Volumes given below are for up to  $10^7$  nucleated cells. When working with fewer than  $10^7$  cells, use the same volumes as indicated. When working with higher cell numbers, scale up all reagent volumes and total volumes, accordingly (e.g. for  $2 \times 10^7$  nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

1. Determine cell number.
2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to  $10^7$  nucleated cells per 100  $\mu$ L of buffer.
4. Add 10  $\mu$ L of the Anti-Ter-119 antibody.
5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2–8 °C).
  - ▲ Note: Working on ice requires increased incubation times. Higher temperatures and/or longer incubation times may lead to non-specific cell labeling.
6. Wash cells by adding 1–2 mL of buffer per  $10^7$  cells and centrifuge at 300×g for 10 minutes. Aspirate supernatant completely.
7. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

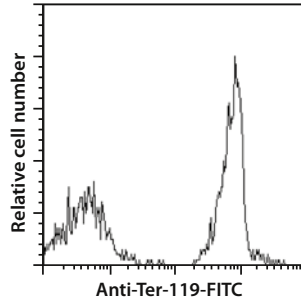
140-001-206.02



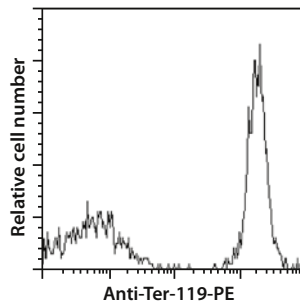
### 3. Examples of immunofluorescent staining with Anti-Ter-119 antibodies

Mouse spleen cells were stained with Anti-Ter-119 antibodies conjugated to FITC (a), PE (b), or APC (c), and analyzed by flow cytometry. Cell debris and dead cells were excluded from the analysis based on scatter signals and PI fluorescence.

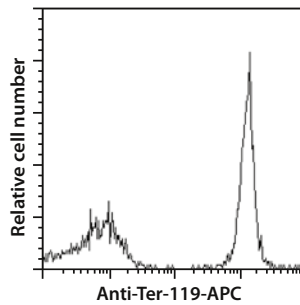
(a) Mouse spleen cells stained with Anti-Ter-119-FITC.



(b) Mouse spleen cells stained with Anti-Ter-119-PE.



(c) Mouse spleen cells stained with Anti-Ter-119-APC.



### 4. References

1. Kina, T. *et al.* (2000) The monoclonal antibody TER-119 recognizes a molecule associated with glycophorin A and specifically marks the late stages of murine erythroid lineage Br J Haematol. 109: 280–287.
2. Ogawa, M. *et al.* (1991) Expression and Function of c-kit in Hematopoietic Progenitor Cells. J. Exp. Med. 174: 63–71.
3. Ikuta, K. *et al.* (1990) A Developmental Switch in Thymic Lymphocyte Maturation Potential Occurs at the Level of Hematopoietic Stem Cells. Cell 62: 863–874.

All protocols and data sheets are available at [www.miltenyibiotec.com](http://www.miltenyibiotec.com).

#### Warnings

Reagents contain sodium azide. Under acidic conditions sodium azide yields hydrazoic acid, which is extremely toxic. Azide compounds should be diluted with running water before discarding. These precautions are recommended to avoid deposits in plumbing where explosive conditions may develop.

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