



Anti-Ig λ light chain antibodies human

Anti-Ig λ light chain-FITC	130-093-040
Anti-Ig λ light chain-PE	130-093-039
Anti-Ig λ light chain-APC	130-093-038
Anti-Ig λ light chain-Biotin	130-093-025
Anti-Ig λ light chain pure	130-093-024

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-Ig λ light chain antibodies

1. Description

Components	1 mL Anti-Ig λ light chain antibodies, human: monoclonal Anti-Ig λ light chain antibodies conjugated to fluorescein isothiocyanate (FITC), R-phycoerythrin (PE), allophycocyanin (APC), or biotin. The unconjugated (pure) antibody is supplied at a concentration of 100 μ g/mL.
Clone	IS7-24C7 (isotype: mouse IgG1).
Capacity	100 tests or up to 10^9 total cells.
Product format	Antibodies are supplied in buffer containing stabilizer and 0.05% sodium azide.
Storage	Store protected from light at 2–8 °C. Do not freeze. The expiration date is indicated on the vial label.

1.1 Background information

Immunoglobulins produced by B cells are composed of two heavy and two light chains, connected by disulfide bonds. Differences in the composition of the heavy chain constant region determine the different isotypes. The light chains of all isotypes are either of κ or λ type.

Clone IS7-24C7 detects human Ig light chains of the λ type. Heavy chains and light chains of the κ type are not detected.

1.2 Applications

- Identification of B cells expressing immunoglobulins of the λ light chain type by flow cytometry or fluorescence microscopy.
- Staining of intracellular immunoglobulins of the λ light chain type in B cells or plasma cells.

1.3 Recommended antibody dilution

For antibody labeling of human cells.

Anti-Ig λ light chain conjugate	FITC	PE	APC	Biotin
Flow cytometry^a				
- In general	1:11	1:11	1:11	1:11
- Formaldehyde-fixed cells	1:11	1:11	1:11	1:11

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

- Cross-reactivity: The Anti-Ig λ light chain antibody is tested to react with rhesus monkey (*Macaca mulatta*) and cynomolgus monkey (*Macaca fascicularis*) cells.

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 human serum albumin, human serum, or fetal bovine serum. Buffers or media containing Ca^{2+} or Mg^{2+} are not recommended for use.
- (Optional) FcR Blocking Reagent, human (# 130-059-901) to avoid Fc receptor-mediated antibody labeling.
- (Optional) Anti-Biotin-FITC (# 130-090-857), Anti-Biotin-PE (# 130-090-756), or Anti-Biotin-APC (# 130-090-856) as secondary antibody reagent in combination with Anti-Ig λ light chain-Biotin.
- (Optional) Inside Stain Kit (# 130-090-477) for intracellular staining.
- (Optional) Anti-FITC MicroBeads (# 130-048-701), Anti-PE MicroBeads (# 130-048-801), Anti-APC MicroBeads (# 130-090-855), or Anti-Biotin MicroBeads (# 130-090-485).
- (Optional) CD19-FITC (# 130-091-328), CD19-PE (# 130-091-247), CD19-APC (# 130-091-248).
- (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 2×10^7 nucleated cells, use twice the volume of all indicated reagent volumes and total volumes).

14-002-0005-01



1. Determine cell number.
2. Centrifuge cell suspension at 300×g for 10 minutes. Aspirate supernatant completely.
3. Resuspend up to 10⁷ nucleated cells per 100 μL of buffer.
4. Add 10 μL of the Anti-Ig λ light chain antibody.
5. Mix well and incubate for 10 minutes in the dark in the refrigerator (2–8 °C).

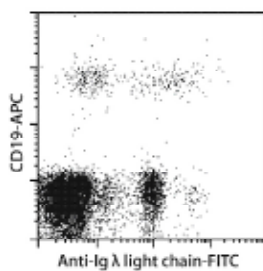
▲ **Note:** Working on ice requires increased incubation times. Higher temperatures and/or longer incubation times may lead to non-specific cell labeling.

6. Wash cells by adding 1–2 mL of buffer per 10⁷ cells and centrifuge at 300×g for 10 minutes. Aspirate supernatant completely.
7. (Optional) If Anti-Ig λ light chain-Biotin was used, resuspend the cell pellet in 100 μL of buffer, add 10 μL of anti-biotin antibody (Anti-Biotin-FITC, Anti-Biotin-PE, or Anti-Biotin-APC), and continue as described in steps 5 and 6.
8. Resuspend cell pellet in a suitable amount of buffer for analysis by flow cytometry or fluorescence microscopy.

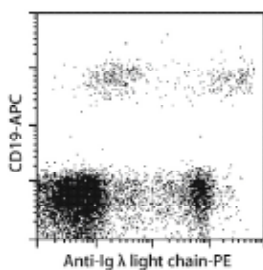
3. Examples of immunofluorescent staining with Anti-Ig λ light chain antibodies

Human peripheral blood mononuclear cells (PBMCs) were stained with Anti-Ig λ light chain antibodies conjugated to FITC (a), PE (b), or APC (c), and analyzed by flow cytometry. Anti-Ig λ light chain -FITC, -PE, and -APC were additionally stained with CD19-PE or CD19-APC. Cells stained with Anti-Ig λ light chain-Biotin (d) were stained with Anti-Biotin-FITC (# 130-090-857) as well as CD19-PE. Cell debris and dead cells were excluded from the analysis based on scatter signals and PI fluorescence.

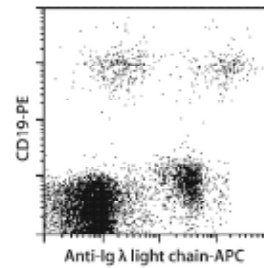
- (a) Human PBMCs stained with Anti-Ig λ light chain-FITC and CD19-APC.



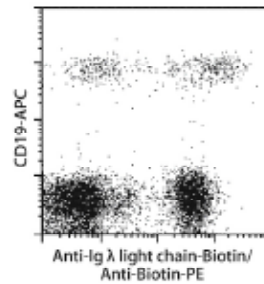
- (b) Human PBMCs stained with Anti-Ig λ light chain-PE and CD19-APC.



- (c) Human PBMCs stained with Anti-Ig λ light chain-APC and CD19-PE.

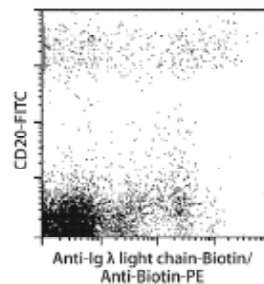


- (d) Human PBMCs stained with Anti-Ig λ light chain-Biotin, Anti-Biotin-APC, and CD19-PE.



Rhesus monkey PBMCs were stained with Anti-Ig λ light chain-Biotin, Anti-Biotin-PE, and CD20-FITC (e) and analyzed by flow cytometry. Cell debris and dead cells were excluded from the analysis based on scatter signals and PI fluorescence.

- (e) Rhesus monkey PBMCs were stained with Anti-Ig λ light chain-Biotin, Anti-Biotin-PE, and CD20-FITC



All protocols and data sheets are available at 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.

Warranty

The products sold hereunder are warranted only to be free from defects in workmanship and material at the time of delivery to the customer. Miltenyi Biotec GmbH makes no warranty or representation, either expressed or implied, with respect to the fitness of a product for a particular purpose. There are no warranties, expressed or implied, which extend beyond the technical specifications of the products. Miltenyi Biotec GmbH's liability is limited to either replacement of the products or refund of the purchase price. Miltenyi Biotec GmbH is not liable for any property damage, personal injury or economic loss caused by the product.

MACS is a registered trademark and autoMACS is a trademark of Miltenyi Biotec GmbH.

© 2007 Miltenyi Biotec GmbH.