MONOCLONAL ANTIBODY CD19

Cat. No.	Form	Quantity	Presentation
1313	Purified	0.2 mg	Freeze-dried
1283	Purified	100 tests	Liquid 2 mL
1284	FITC	100 tests	Liquid 2 mL
1285	PE	100 tests	Liquid 2 mL
1500	PE-Cy5	100 tests	Liquid 2 mL
2470	APC	100 tests	Liquid 1 mL

Warning ***APC-conjugated forms of the IOTest® line of reagents are to be used at 10 µL / test instead of 20 µL / test.***

Clone J4.119

Isotype IgG1 (mouse)

Immunogen SKLY 18 lymphoma cells

Hybridoma NS1 x Balb/c spleen cells

Specificity The CD19 antigen is a single chain, type I integral membrane glycoprotein with a molecular weight of 95 kDa (1). Expression of the CD19 molecule is found on all B lymphocytes, including pro-B cells, but is lost during maturation to plasma cells (2-4). The CD19 antigen is also expressed on the membrane of follicular dendritic cells and on most stabilized B cell lines.

In adults about 15% of peripheral blood lymphocytes are $CD19^+$. This percentage is lower during early childhood and reaches adult values at approximately 6 years of age (5, 6). CD19 expression is not observed in normal T lymphocytes, NK cells, monocytes, and granulocytes (2).

The CD19 molecule appears important in regulating B-cell activation and proliferation (7).

In vitro studies show that the CD19 antibody has an inhibitory effect on the activation and proliferation of B lymphocytes.

The J4.119 monoclonal antibody was assigned to the CD19 cluster of differentiation at the IVth International Workshop on Human Leucocyte Differentiation Antigens in Oxford (1986) (1).

Applications Flow cytometry:
Studies of CD19-expressing lymphocytes.

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Buffer

Freeze-dried form: 1 mg/mL of bovine serum albumin in phosphate-buffered saline.
Liquid forms: 2 mg/mL of bovine serum albumin in phosphate-buffered saline containing 0.1% sodium azide.

Conjugation

FITC: Fluorescein isothiocyanate (FITC) is conjugated at 4-8 moles of FITC per mole of IgG.
Excitation wavelength: 488 nm
Maximum emission wavelength: 525 nm
Main emission color: Green

PE: R-phycoerythrin (PE) is conjugated at 0.7-1 mole of PE per mole of IgG.
Excitation wavelength: 488 nm
Maximum emission wavelength: 575 nm
Main emission color: Orange-red

PE-Cy5: The IgG is conjugated to a tandem dye constituted of R-phycoerythrin covalently linked to cyanin 5.1 at 0.7-1 mole of PE-Cy5 per mole of IgG.
Excitation wavelength: 488 nm
Maximum emission wavelength: 670 nm
Main emission color: Deep-red

APC: Allophycocyanin (APC) is conjugated at 0.7-1 mole of APC per mole of IgG.
Excitation wavelength: 633-635 nm
Maximum emission wavelength: 660 nm
Main emission color: Deep-red

Limitation: APC conjugates are recommended for use only on flow cytometers equipped with an exciting source of 633 nm (He-Ne laser) or 635 nm (Red diode laser).

Reconstitution and Storage

The freeze-dried form may be stored at 2-8°C until the expiration date stated on the vial label. Reconstitute with 1 mL of distilled water. No preservative has been added. The reconstituted form may be stored at -20°C until the expiration date. Aliquotting is suggested to avoid multiple freeze-thaw cycles. The addition of sodium azide at 0.1% (w/v) is recommended for storage of the reconstituted form for up to one month at 2-8°C.

The purified liquid form should be stored at 2-8°C until the expiration date stated on the vial label.

The conjugated forms should not be frozen and should be stored in the dark at 2-8°C until the expiration date stated on the vial label.

Recommended Procedures

Flow cytometry

Purified, FITC-, PE- and PE-Cy5-conjugated forms: 20 µL / 5 x 10^5 cells or 100 µL whole blood.

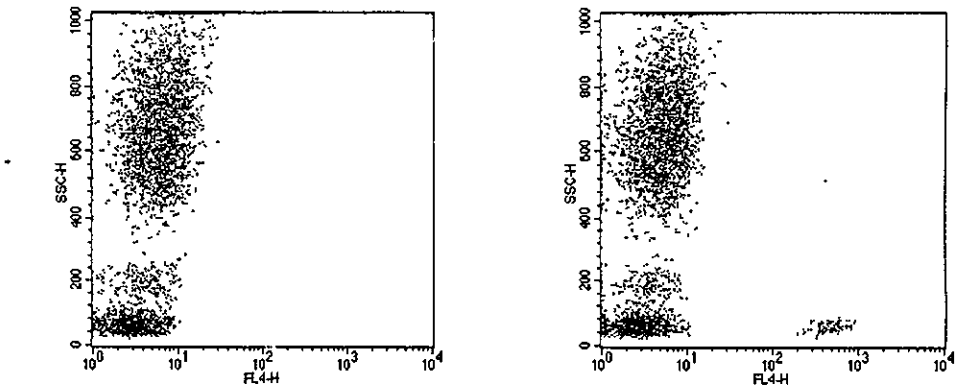
APC-conjugated form: A specific calibration is applied to facilitate the blending of conjugated antibodies in multiparametric flow cytometry.
10 µL / 5 x 10^5 cells or 100 µL whole blood.

Limitation: R-phycoerythrin (PE) is sensitive to light exposure. Consequently, PE- or PE-Cy5-conjugated antibodies are not suitable for fluorescence microscopy.

Cat. No. 1313

Results Example

The graphs below are double parameter representations (Side Scatter *versus* Fluorescence 4) of a lyzed whole blood sample from an healthy donor. Staining is with IgG1-APC (left) and CD19-APC (right). Along the Y axis, lymphocytes are events with low side scatter values, monocytes show low to medium side scatter values and neutrophils show medium to high side scatter values.



Isotypic control, IgG1-APC (Cat. No 2475) Specific staining, CD19-APC (Cat. No 2470)

Analysis is with a Becton Dickinson FACSCalibur™ flow cytometer equipped with CELLQuest™ Software.

References

1) Tedder, F., Isaacs, C.M., Penta, A., "Cloning and structure of CD19, a member of the immunoglobulin superfamily. Use of transfected celles to examine the Workshop antibodies", 1989, in Leucocyte Typing IV, White Cell Differentiation Antigens, Knapp, W., et al., Eds., Oxford Univ. Press, p. 36-38.

2) Zhou, L.J., Tedder, T., "CD19 Workshop Panel report", 1995, in Leucocyte Typing V, White Cell Differentiation Antigens, Schlossman, S.F., et al., Eds Oxford Univ. Press, p. 507-509.

3) Uckun, F.M., "Regulation of human B-Cell ontogeny", 1990, Blood, **76**, 10, 1908-1923.

4) Loken, M.R., Shah, V.O., Dattilio, K.L., Civin, C.I., "Flow cytometric analysis of human bone marrow. II. Normal B lymphocyte development", 1987, Blood, **70**, 5, 1316.

5) Caldwell, C.W., Poje, E., Helikson, M.A., "B-cell precursors in normal pediatric bone marrow", 1991, Am. J. Clin. Pathol., **95**, 816.

6) Hannet, I., Erkeller-Yuksel, F., Lydyard, P., Deneys, V., DeBruyère, M., "Developmental and maturational changes in human blood lymphocyte subpopulations", 1992, Immunol. Today, **13**, 6, 215-218.

7) Carter, R.H., Doody, G.M., Bolen, J.B., Fearon, D.D., "Membrane IgM-induced tyrosine phosphorylation of CD19 requires a CD19 domain that mediated association with components of the B cell antigen receptor complex", 1997, J. Immunol., **158**, 3062-3069.

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