IOTest® IgG1-FITC / IgG2a-PE

PN A10973 - 1 mL liquid - 20 µL / test

For Research Use Only. Not For Use In Diagnostic Procedures.

REAGENT
IOTest Conjugated Antibodies – IgG1-FITC / IgG2a-PE
PN A10973 – 1 mL liquid – 20 µL / test*

	CLONE 1	CLONE 2
Specificity	IgG1	IgG2a
Clone	679.1Mc7	7T4-1F5
Hybridoma	P3-X63-Ag.8.653 x Balb/c	NS1 x Balb/c
Ig Chain	IgG1	IgG2a
Species	Mouse	Mouse
Source	Ascites fluid	Ascites fluid
Purification	Affinity chromatography on Protein A	Affinity chromatography on Protein A
Conjugation	FITC (Fluorescein isothiocyanate)	PE (R-Phycoerythrin)
Molar Ratio	FITC / Ig: 2 – 5	PE / Ig: 0.5 – 1.5

SPECIFICITY
679.1Mc7 and 7T4-1F5 are mouse monoclonal antibodies of the IgG1 and IgG2a isotype, respectively. 679.1Mc7 is FITC-conjugated and 7T4-1F5 is PE-conjugated. They have an irrelevant specificity and induce nonspecific immunolabeling on cells and therefore constitute isotype controls. When using a specific monoclonal antibody for a flow cytometric analysis, the level of nonspecific antibody binding in cell surface staining must be monitored to identify positive cells (1). This can be done by using a monoclonal antibody devoid of any relevant specificity with regards to the studied cell population, providing that it shares the same characteristics (species, isotype and conjugated fluorochrome) with the monoclonal antibody of interest (2).

BUFFER
2 mg/mL bovine serum albumin in phosphate-buffered saline containing 0.1 % sodium azide.

CONJUGATION
Fluorescein isothiocyanate (FITC) is conjugated at 2 – 5 moles of FITC per mole of Ig.
Excitation wavelength: 488 nm
Maximum emission wavelength: 525 nm
Main emission color: Green

R-phycoerythrin (PE) is conjugated at 0.5 – 1.5 moles of PE per mole of Ig.
Excitation wavelength: 488 nm
Maximum emission wavelength: 575 nm
Main emission color: Orange-red

APPLICATION
This dual color reagent (IgG1-FITC / IgG2a-PE) is used to monitor the level of nonspecific staining in cell staining procedures which use antibody mixtures of the mouse IgG1 and IgG2a subclasses conjugated to FITC and PE, respectively. Its formulation is optimized for cell surface immunolabeling studies.

STATEMENT OF WARNINGS
1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.
2. Specimens, samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
3. Never pipet by mouth and avoid contact of samples with skin and mucous membranes.
4. Do not use reagent beyond the expiration date on the vial label.
5. Minimize exposure of reagent to light during storage or incubation.
6. Avoid microbial contamination of reagent or erroneous results may occur.
7. Use Good Laboratory Practices (GLP) when handling this reagent.

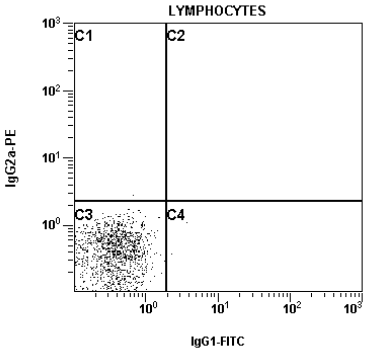
STORAGE CONDITIONS AND STABILITY
This reagent is stable to the expiration date when stored at 2 – 8°C. Do not freeze. Minimize exposure to light.

REAGENT PREPARATION
No reconstitution is necessary. This reagent may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

PROCEDURE
This reagent is designed for flow cytometry.
Assay volume: 20 µL per 5 x 10^5 cells in one test, or per 100 µL whole blood. A wash is required to yield optimal results.

EXAMPLE DATA
The graph below is a biparametric representation (fluorescence intensity versus fluorescence intensity) of lyzed normal whole blood sample. Staining is with IgG1-FITC / IgG2a-PE isotypic control (PN A10973) gated on lymphocytes.

Acquisition is with a COULTER (R) EPICS (R) XL (TM) flow cytometer.
Analysis is with the CYTOMICS (TM) RXP (TM) analysis software.



SELECTED RESEARCH REFERENCES

1. MMWR, "1997 revised guidelines for performing CD4+ T-cell determinations in persons infected with human immunodeficiency virus (HIV)", 1997, INIST CNRS, RR-2, 46, 1-29.
2. Stewart, C.C., Stewart, S.J., "Cell preparation for the identification of leukocytes", 1994, Methods in cell biology, Chap3, 41, 39-60.

PRODUCT AVAILABILITY
IOTest IgG1-FITC / IgG2a-PE
Conjugated Antibodies
PN A10973 – 50 tests – 20 µL/test*

TRADEMARKS

For additional information in the USA, call 800-526-7694.

Manufactured by:
Immunotech, a Beckman Coulter Company
130, avenue de Lattre de Tassigny, B.P. 177
13276 Marseille Cedex 9, France
www.beckmancoulter.com

Insert No.: A10973EX_A 2003-10-17
AC-03-0934

BECKMAN COULTER™