

**CYTO-STAT®
triCHROME™
CD45-FITC/
MslgG1-RD1/
MslgG1-PC5**

REF 6607019 - 50 tests

PN 4238017-F



	CLONE 1	CLONE 2	CLONE 3
Specificity	CD45	Non Applicable	Non Applicable
Clone	B3821F4A	2T8-2F5	2T8-2F5
Hybridoma	NS-1 x BALB/c	NS-1 x BALB/c	NS-1 x BALB/c
Immunogen	Transfectant containing cDNA of human CD45	Non Applicable	Non Applicable
Ig Chain	IgG2b	IgG1	IgG1
Species	Mouse	Mouse	Mouse
Source	Ascites fluid	Conditioned media	Conditioned media
Purification	Affinity chromatography	Affinity chromatography	Affinity chromatography
Fluorescence	Excites at 468-509 nm / Emits at 504-541 nm	Excites at 486-580 nm / Emits at 568-590 nm	Excites at 486-580 nm / Emits at 660-680 nm
Conjugation	FITC (Fluorescein Isothiocyanate)	RD1 (Phycoerythrin)	PC5 (Phycoerythrin-Cy5)
Molar Ratio	FITC/Protein: 3-10	RD1/Protein: 0.5-1.5	PC5/Protein: 0.5-1.5

ISOTYPIC CONTROL

For In Vitro Diagnostic Use

INTENDED USE

CYTO-STAT triCHROME CD45-FITC/MslgG1-RD1/MslgG1-PC5 Isotypic Control is a three-color fluorescent reagent comprised of three murine monoclonal antibodies. Each antibody is labeled with a different fluorochrome. This product is intended for use as a quality control reagent to monitor the levels of nonspecific antibody binding in cell surface staining procedures which use CYTO-STAT triCHROME Monoclonal Antibody Reagents comprised of CD45-FITC and two IgG1 subclass Monoclonal Antibodies conjugated to RD1 and PC5.

Refer to the specific CYTO-STAT triCHROME Monoclonal Antibody Reagent package insert for instructions on how to use this Isotypic Control reagent.

ANTIBODY SPECIFICITY

The CD45 antibody recognizes members of the CD45 family of pan leukocyte antigens with molecular weights of 180, 190, 210 and 220 kD.¹⁻³ It is also known as the leukocyte common antigen (LCA). CD45 antigen is expressed on every type of hematopoietic cell except mature erythrocytes and their immediate progenitors.^{4,5} It has not been detected in differentiated nonhematopoietic tissue.^{4,7}

The MslgG1 antibodies are used to monitor the levels of nonspecific staining in cell staining procedures which use antibodies of the mouse IgG1 subclass conjugated to RD1 and PC5.⁸⁻¹⁰

REAGENT

CYTO-STAT triCHROME CD45-FITC/MslgG1-RD1/MslgG1-PC5
PN 6607019 - 50 tests (0.5 mL)

CLONE: B3821F4A (CD45) was derived from the hybridization of mouse NS-1 myeloma cells with spleen cells from BALB/c mice immunized with a transfectant containing cDNA of human CD45.¹¹
2T8-2F5 (MslgG1) was derived from the hybridization of mouse NS-1 myeloma cells with spleen cells from BALB/c mice.

Ig CHAIN: Mouse IgG2b (CD45) heavy chain and kappa light chains.
Mouse IgG1 (MslgG1) heavy chain and kappa light chains.

CYTOTOXICITY: Complement-mediated lysis (CD45).
None by direct lysis (MslgG1).

SOURCE: Ascites fluid (CD45)
Conditioned media (MslgG1)

PURIFICATION: Affinity chromatography

CONJUGATION: CD45-FITC (Fluorescein isothiocyanate)
MslgG1-RD1 (Phycoerythrin)
MslgG1-PC5 (Phycoerythrin-Cy5)

MOLAR RATIO: FITC/protein 3-10
RD1/protein 0.5-1.5
PC5/protein 0.5-1.5

FLUORESCENCE:
FITC (Green) Excites at 468-509 nm
Emits at 504-541 nm
RD1 (Orange) Excites at 486-580 nm
Emits at 568-590 nm
PC5 (Deep Red) Excites at 486-580 nm
Emits at 660-680 nm

REAGENT CONTENTS

The concentration of nonantibody reagents is 0.2% BSA, 0.01 M potassium phosphate, 0.15 M NaCl, 0.1% NaN₃ and stabilizers.

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. This reagent is designed for use with whole blood preparations. It may be used with the lyophilized lymphocyte preparation COULTER® CYTO-TROL™ Control Cells.
7. Avoid microbial contamination of reagent or erroneous results may occur.
8. Use Good Laboratory Practices (GLP) when handling this reagent.
9. Review all histograms before reporting results.
10. Harmful if swallowed.
11. After contact with skin, wash immediately with plenty of water.

STORAGE CONDITIONS AND STABILITY

Unopened reagent is stable to the expiration date on the vial label when stored at 2-8°C. Opened vials are stable for 90 days when stored at 2-8°C. Return reagents to 2-8°C immediately after use. Do not freeze. Minimize exposure to light.

EVIDENCE OF DETERIORATION

Any change in the physical appearance of this reagent (normal appearance is a clear, pink liquid) or any major variation in values obtained for control samples may indicate deterioration and the reagent should not be used.

REAGENT PREPARATION

No preparation is necessary. This CYTO-STAT triCHROME Isotypic Control reagent is used directly from the vial.

USAGE

This CYTO-STAT triCHROME Isotypic Control reagent is for use with standard flow cytometry methodologies using the COULTER® IMMUNOPREP™ Reagent System and Q-PREP™, MULTI-Q-PREP™ or TQ-PREP™ Workstation. It incorporates lymphocyte gating using Side Scatter (SS) and the CD45 bright positive population (see Figure 1). Refer to the specific CYTO-STAT triCHROME Monoclonal Antibody Reagent package insert for instructions on how to use this control reagent.

The following are representative histograms for a whole blood specimen stained with CYTO-STAT triCHROME CD45-FITC/ MslgG1-RD1/ MslgG1-PC5, lysed with COULTER IMMUNOPREP reagents and analyzed using the COULTER EPICS® XL™ flow cytometer.

Figure 1. A two-parameter CD45-FITC vs. SS histogram to identify lymphocytes.

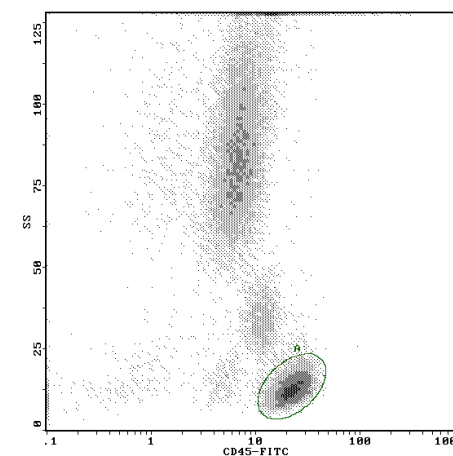
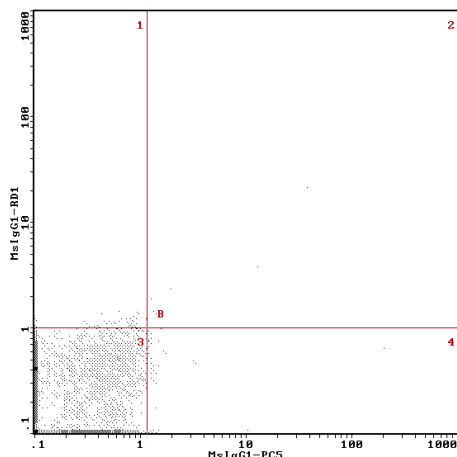


Figure 2. A two-parameter MslgG1-PC5 vs. MslgG1-RD1 histogram gated on lymphocytes used to establish Quad Stats at 98% (nominal) nonspecific staining.



SELECTED RESEARCH REFERENCES

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PRODUCT AVAILABILITY

CYTO-STAT triCHROME CD45-FITC/
MslgG1-RD1/MslgG1-PC5
PN 6607019 - 50 tests (0.5 mL)

RD1 is licensed under patent 4,520,110.
PC5 is licensed under patents 4,520,110 and 4,542,104.
Cy5 is licensed under patents 4,981,977 and 5,268,486.

For additional information in the US call 800-526-7694.
Outside the US, contact your local Coulter Representative.

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