

PN IM3604**TCR V β 7.2-PE****(Zizou4)****IO Test[®]**

Conjugated Antibodies

50 tests**20 μ L / test****For Research Use Only. Not For Use In Diagnostic Procedures.****SPECIFICITY**

Human variable β 7.2 chain of the T-cell receptor (V β 7.2) is also called TCRBV7S2, according to the nomenclature from Wei et al. (1).

The monoclonal antibody Zizou4 recognizes the human TCR V β 7.2 allele product (BV7S2 cDNA, in ref. 2). A genetically determined insertion/deletion related polymorphism in the TCRB locus, including the functional V β 7.2 variable gene segment, has been described (2). Consequently, the expression of the V β 7.2 allele product may be negative for some individuals (V β 7.2 deleted / V β 7.2 deleted haplotypes).

This antibody stains from 0% (V β 7.2 del / V β 7.2 del haplotypes) to 5.5% (mean 1.5%) of peripheral CD3-positive lymphocytes in normal blood (data on file at Immunotech).

REAGENT

Clone	Zizou4
Isotype	IgG2a
Species	Mouse
Immunogen	Mouse T-cell hybridoma 58 α - β -, transfected with the human TCR-V β 7.2 gene
Hybridoma Source	Myeloma X63Ag8 x Balb / c Ascites fluid
Purification	Ion exchange or affinity chromatography
Conjugation	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
Buffer	2 mg/mL bovine serum albumin in phosphate-buffered saline containing 0.1% sodium azide.

APPLICATION

T-cell repertoire studies by flow cytometry.

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 extensively 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 antibody beyond the expiration date on the label.
5. Do not expose reagents to strong light during storage or incubation.
6. Avoid microbial contamination of reagents or incorrect results might occur.

STORAGE CONDITIONS AND STABILITY

Each reagent is stable up to the expiration date when stored at 2 – 8°C. Do not freeze. Minimize exposure to light.

REAGENT PREPARATION

No reconstitution is necessary. This monoclonal antibody 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 μ L per 5 x 10⁶ cells in one test, or per 100 μ L whole blood or bone marrow.

A wash is required to yield optimal results.

Warning: As this antibody recognizes a small population, it is often preferable to use double staining experiments with another conjugated T-cell marker (CD2, CD3, CD4, CD8, etc.).

EXAMPLE DATA

Warning: Due to described polymorphisms (deletion of the V β 7.2 gene segment), some blood samples may be negative for the expression of TCR V β 7.2 (2).

The graphs below are biparametric representations (Fluorescence Intensity versus Fluorescence Intensity) of a lyzed normal, positive whole blood sample. The staining is performed with a combination of 2 reagents, CD3-FITC (PN IM1281) and TCR V β 7.2-PE (PN IM3604). The gating is done on CD3⁺ lymphocytes.

*Upper-right quadrant (2): CD3⁺ - TCR V β 7.2⁺ T lymphocytes.

*Lower-right quadrant (4): CD3⁺ - TCR V β 7.2⁻ T lymphocytes.

Figure 1: Acquisition with a COULTER[®] EPICS[®] XL[™] flow cytometer. Analysis with the XL SYSTEM II[™] software.

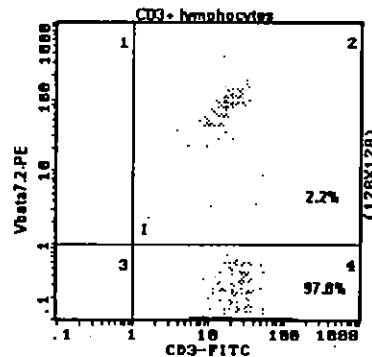
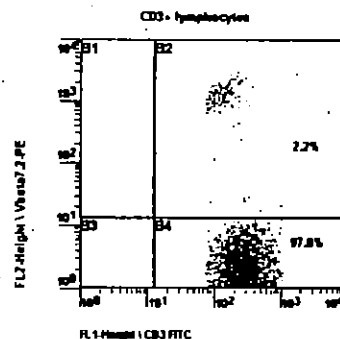


Figure 2: Acquisition with a Becton Dickinson FACScan[™] flow cytometer equipped with the LYSYS II[™] software. Analysis with EXPO[™] Cytometer software (Coulter PN 6605434).



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PARTNERS IN CELL ANALYSIS

**IMMUNOTECH**
A COULTER COMPANY

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50 tests

20 μ L / test

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SELECTED RESEARCH REFERENCES

1. [70] Wei, S., Charmley, P., Robinson, M.A., Concannon, P., "The extent of the human germline T-cell receptor V β gene segment repertoire", 1994, Immunogenetics, 40, 27-36
2. [213] Zhao, T.M., Whitaker, S.E., Robinson, M.A., "A genetically determined insertion / deletion related

polymorphism in human T cell receptor β chain (TCRB) includes functional variable gene segments", 1994, J. Exp. Med., 180, 1405-1414

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