

MONOCLONAL ANTIBODY

Vβ2

Cat. No.	Form	Quantity	Presentation
2006	Purified	0.1 mg	Freeze-dried
2081	Biotin	0.1 mg	Freeze-dried
2213	PE	50 tests	Liquid 1 mL
2407	FITC	50 tests	Liquid 1 mL

Clone MPB2D5

Isotype IgG1 (mouse)

Immunogen Human T-cell line

Hybridoma Myeloma X63 Ag8.653 x SJL spleen cells

Specificity Human variable β2 chain of the T-cell receptor. The antibody recognizes all alleles of the single membered of Vβ2 family (described in (1)) also called TCRBV2S1 according to the nomenclature from Wei et al. (2). This antibody has been further characterized by cell sorting on PBL using this monoclonal antibody followed by analysis of sorted cells by molecular biology (3).

MPB2D5 stains 4.2 to 9.1% of peripheral CD3 positive lymphocytes from 6 healthy adult donors (data on file at Immunotech).

This antibody is described in ref. 3 and has been confirmed at the First Human TcR Monoclonal Antibody Workshop in San Francisco in 1995 (4).

Applications T-cell repertoire studies in normal and pathological situations including autoimmune disease, graft rejection and AIDS (5).

Superantigenic stimulation of T cells ; Vβ2 is for example the target of TSST1 (3, 6).

Buffer Freeze-dried forms: 1 mg/mL bovine serum albumin in phosphate buffered saline.

Liquid form: 2 mg/mL bovine serum albumin in phosphate buffered saline containing 0.1% sodium azide.

Reconstitution and Storage The freeze-dried form may be stored at 2 - 8 °C until the expiration date. Reconstitute with 0.5 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.

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MA003

FOR RESEARCH USE ONLY - NOT FOR USE IN DIAGNOSTIC PROCEDURES



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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

Freeze-dried forms: 2 µg / 5×10^5 cells / test or 100 µL whole blood.

Liquid form: 20 µL / 5×10^5 cells / test or 100 µL whole blood.

Since this antibody recognizes a small cell population, it is often preferable to use double staining experiments with another T-cell marker (CD2, CD3, CD4, CD8, etc.). Double staining is also possible with the purified unlabelled form using the following protocol.

A. Double labelling protocol using freeze-dried unconjugated form (Cat. No. 2006) with CD3 PE (Cat. No. 1282)

- 1 To 100 µL of whole blood, add 10 µL of the reconstituted purified antibody. Incubate 15 minutes at room temperature (RT).
2. Add 3 mL of PBS/BSA/NaN₃. Centrifuge 5 minutes 1200 rpm, discard supernatant.
3. Add 100 µL of secondary antibody F(ab')₂ goat anti-mouse Ig conjugated to FITC at recommended dilution in PBS/BSA/NaN₃. Incubate 15 minutes at room temperature.
4. Wash, by repeating step 2.
5. Resuspend cells in 100 µL of PBS/BSA/NaN₃ containing 1 mg/mL of total mouse Ig (to saturate eventual free sites of the goat anti-mouse FITC). Incubate 5 minutes at room temperature.
6. Without washing, add 20 µL of CD3 PE and incubate 15 minutes at room temperature.
7. Wash, by repeating step 2.
8. Proceed as usual for lysis of red blood cells and fixing of white cells.

B. Double labelling protocol using biotinylated form. (Cat. No. 2081) with CD3 PE (Cat. No. 1282)

1. To 100 µL of whole blood add 10 µL of the reconstituted biotinylated form, and 20 µL of CD3 PE. Incubate 15 minutes at room temperature.
2. Add 3 mL PBS/BSA/NaN₃. Centrifuge 5 minutes at 1200 rpm, discard supernatant.
3. Add 100 µL of FITC conjugated streptavidin at the recommended dilution.
4. Wash, by repeating step 2.
5. Then proceed as usual for lysis of red blood cells and fixing of white cells.