

PRODUCT INFORMATION SHEET

Monoclonal antibodies detecting human antigens

CD4 FITC **CD25** R-PE RUO REF IQP-260FR 50 tests

RUO *For Research Use Only*



Description

The combination of CD4 and CD25 is used to detect activated CD4⁺ T cells (CD25⁺) and regulatory CD4⁺ T cells (CD25⁺).

CD4

Clone Edu-2

Isotype Murine IgG2b

Specificity Monoclonal antibodies clustered as CD4 detect most thymocytes and a subpopulation of peripheral blood T cells, called T helper cells (Th). In addition, CD4 is expressed on monocytes and macrophages. The CD4 antigen is a 55 kD glycoprotein which plays a role in the recognition of foreign antigens presented to T cells by MHC class II molecules. Furthermore, CD4 acts as a receptor for HIV-1 by binding the viral protein gp120.

HLDA Workshop

5th International Workshop on Human Leukocyte Differentiation Antigens

CD25

Clone B-B10

Isotype Murine IgG1

Specificity CD25 is the α-chain of the Interleukin-2 (IL-2) receptor. The IL-2 receptor is expressed on activated cells including T cells, B cells and monocytes. It is also present on a subset of thymocytes, HTLV-1 transformed T and B cells, EBV transformed B cells, myeloid precursors and oligodendrocytes. IL-2 induces the expression of the CD25 subunit on NK cells.

HLDA Workshop

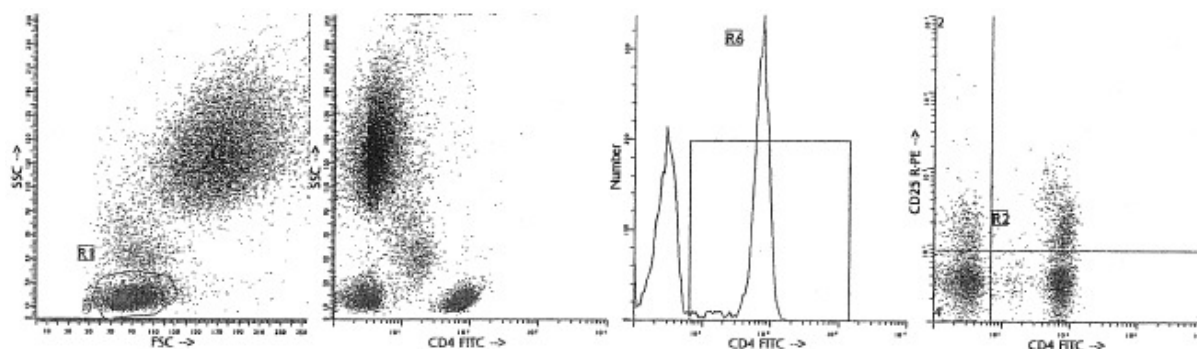
4th International Workshop on Human Leukocyte Differentiation Antigens

Usage

All these reagents are effectively formulated for direct immunofluorescent staining of human tissue for flow cytometric analysis using 10 μl/10⁶ leukocytes for singles and 20 μl/10⁶ leukocytes in case of dual and triple combinations. Since applications vary, each investigator should titrate the reagent to obtain optimal results.

Representative Data

Staining with the dual of CD4 FITC and CD25 R-PE and analysis by flow cytometry is illustrated. Direct staining was performed using 20 μl of the conjugated monoclonal antibody preparation and 100 μl of blood sample.



Immunofluorescence staining and lysing protocol

Flow cytometry method for use with dual and triple combinations

1. Add 100 µl of EDTA-treated blood (i.e. approx. 10^6 leukocytes) to a 5 ml reagent tube. The content of one tube is sufficient to perform one test.
For combinations with anti-kappa and/or anti-lambda Ig see application note below.
2. Add to each tube 20 µl of labeled monoclonal antibody combination*.
3. Vortex the tube to ensure thorough mixing of antibody and cells.
4. Incubate the tube for 15 minutes at room temperature in the dark.
5. Add 100 µl of IQ Lyse (IQP-199 ready-to-use) and mix immediately.
6. Incubate for 10 minutes at room temperature in the dark.
7. Add 2 ml of demineralized water and incubate for 10 minutes in the dark.
8. Centrifuge the labeled cell suspension for 2 minutes at 1000 x g.
9. Remove the supernatant and resuspend the cells in 200 µl of PBS**.
10. Analyze by flow cytometry within four hours (alternatively, the cells may be fixed by 0.05% of formaline in buffered saline for analysis the next day. Some antigens are readily destroyed upon fixation and this should be taken into account when using this alternative).

* Appropriate mouse Ig isotype control samples can be included in any labeling study

** PBS: Phosphate Buffered Saline, pH 7.2

Application note for anti-kappa and/or anti-lambda Ig combinations

Add 2 ml of PBS containing 0.001% (v/v) Heparin (**prewarmed to 37 °C**) to the cell suspension

Vortex, centrifuge (2 min at 300x g) and discard the supernatant

Repeat this step twice

Resuspend the pelleted blood cells in 100 µl PBS containing 0.001% (v/v) Heparin



Handling and Storage

Antibodies are supplied either as 100 tests per vial (1 ml) for singles or 50 tests per vial (1 ml) for dual and triple combinations. They are supplied in 0.01 M sodium phosphate, 0.15 M NaCl; pH 7.3, 0.2% BSA, 0.09% sodiumazide (NaN_3). Store the vials at 2-8 °C. Monoclonal antibodies should be protected from prolonged exposure to light. Reagents are stable for the period shown on the vial label when stored properly.








Warranty Products sold hereunder are warranted only to conform to the quantity and contents stated on the label at the time of delivery to the customer. There are no warranties, expressed or implied, which extend beyond the description on the label of the product. IQ Products is not liable for property damage, personal injury, or economic loss caused by the product.

Characterization

To ensure consistently high-quality reagents, each batch of monoclonal antibody is tested for conformance with characteristics of a standard reagent. Representative flow cytometric data is included in this data sheet.

Warning All products contain sodiumazide. This chemical is poisonous and hazardous. Handling should be done by trained staff only.

Explanation of used symbols


| | |
|---|---|
|  | Consult instructions for use |
|  | Catalogue number |
|  | Sufficient for |
|  | In Vitro Diagnostic medical device |
|  | Caution, consult accompanying document |
|  | Keep away from (sun)light |
|  | Biological risks |
|  | Temperature limitation (°C) |
|  | For Research Use Only |
|  | Batch code |
|  | Use by yyyy-mm-dd |
|  | Manufacturer |
|  | Authorized Representative in the European Community |
|  | Conformité Européenne (European Conformity) |


| | | Label - tandem | Ex -max (nm) | Em -max (nm) |
|-----|-------------|-----------------------|---------------------|---------------------|
| P | PURE | purified material | - | - |
| F | FITC | FITC | 488 | 519 |
| R | R-PE | PE | 488, 532 | 578 |
| C | CyQ | PE-Cy5.18 | 488, 532 | 667 |
| A | APC | | 595, 633, 635, 647 | 660 |
| PC | PerCP | | 488, 532 | 678 |
| PCC | PerCP-Cy5.5 | | 488, 532 | 695 |



IQ Products BV

Rozenburglaan 13a
9727 DL Groningen, The Netherlands


 +31 (0)50 57 57 000

 +31 (0)50 57 57 002

 Technical marketing@iqproducts.nl

 Orders orders@iqproducts.nl

 www.iqproducts.nl

 **Products**
bright fluorescence