# Anti-Human CD45RA (RP1/11)

| | REF | Σ | | [A] | |
|---|---|---|---|---|---|
| PURE | 45RAPU | 1 mg | 1 mg/ml | | RUO |
| FITC | 45RAF-100T | 100 test | 20 µL/test | 2 mg/ml | |
| PE-Cyanine7 | 45RAPC7-100T | 100 test | 3 µL/test | 0,03 mg/ml | |
| APC | 45RAA-100T | 100 test | 5 µL/test | 0,05 mg/ml | |

## 1. PRODUCT DESCRIPTION

**Clone:** RP1/11;
**Isotype**: Mouse IgG1;
**Tested application**: flow cytometry;
**Immunogen**: The anti-CD45RA monoclonal antibody derives from immunizing mice with purified human CD45;
**Species reactivity**: Human;
**Storage instruction:** store in the dark at 2-8 °C;
**Storage buffer:** aqueous buffered solution containing protein stabilizer and 0.09% sodium azide ($NaN_3$);
**Recommended usage**: Immunostep's CD45RA, clone RP1/11 is a monoclonal antibody intended for the identification and enumeration of helper/inducer T-cell subset and CD45RA+ cells using flow cytometry. This reagent is effective for direct immunofluorescence staining of human tissue for flow cytometric analysis using 1 test for $10^6$ cells;
**Presentation**: liquid;
**Source**: Supernatant proceeding from an in vitro cell culture of a cell hybridoma;
**Purification**: Affinity chromatography;
**Other names**: Ptprc, B220, CD45, CD45R, GP180, L-CA, LY5, T200;
**Gene ID:** 5788;
**Molecular weight:** 220 kDa.

## 2. ANTIGEN DETAILS

**Large description:** The RP1/11 monoclonal antibody reacts with human CD45RA, a 220 kDa molecule expressed by subpopulations of CD4+ peripheral T lymphocytes, CD8+ peripheral T lymphocytes, and B cells. The CD45RA+ T cell populations are mainly naive/virgin allowing the use of RP1/11 mAb as a phenotypic marker to discriminate T cell subsets.[1-6]

## 3. WARRANTY

Warranted only to conform to the quantity and contents stated on the label or in the product labelling at the time of delivery to the customer. Immunostep disclaims hereby other warranties.

Immunostep's sole liability is limited to either the replacement of the products or refund of the purchase price.

## 4. ADDITIONAL INFORMATION

For research use only. Not for diagnostic use.

Not for resale. Immunostep will not be responsible of violations that may occur with the use of this product. Any use of this product other than the specified in this document is strictly prohibited.

Unless otherwise indicated by Immunostep by written authorization, this product is intended for research only and is not to be used for any other purpose, including without limitation, for human or animal diagnostic, therapeutic or commercial purposes.

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## 5. PROTOCOL

**Direct Immunofluorescence Cell Surface Staining Protocol**

1. Transfer 100 ul (106 cells/test) of the sample to a 12 x 75 mm polystyrene test tube.
2. Add the suggested volume indicated on the antibody vial to the 12x75 mm cytometer tube.
3. Mix well and incubate in the dark at room temperature at 4 °C for 30 minutes or at room temperature (20-25 °C) for 15 minutes.
4. After the incubation period, add 1,5 ml of an erythrocyte-lysing solution and mix. Incubate at room temperature in the darkness (the blood should be well mixed with the lysing solution).
5. Centrifuge tubes at 540xg for 5 minutes. The supernatant is removed with a Pasteur pipette or with a vacuum pump.
6. Resuspend and wash with 3-5 mL of PBS at 540xg for 5 min.
7. After removing the supernatant and resuspending the cell pellet, add 300 µL of PBS and adquire on the flow cytometer are recorded.
8. Analyse on a flow cytometer or store at 2- 8 °C in the dark until analysis. Samples can be run up to 24 hours after lysis.

**Indirect Immunofluorescence Cell Surface Staining Protocol**

1. Transfer 100 ul (106 cells/test) of the sample to a 12 x 75 mm polystyrene test tube
2. Add purified reagent according to manufacturer's recommendation and mix gently with a vortex mixer.
3. Incubate in the dark at room temperature at 4 °C for 30 minutes or at room temperature (20-25 °C) for 15 minutes.
4. Add 2 mL 0.01 mol/L PBS (It betters that it containing 2% bovine serum albumin) and resuspend the cells by using a vortex mixer. Centrifuge at 540xg for 5 min in order to remove the McAb not bound to its antigen.
5. Add a secondary conjugated reagent with some fluorochrome and mix. Incubate at room temperature for 15 min in the dark. The absence of light is necessary as the fluorochrome is photoinstability.
6. After the incubation period, add 1,5 ml of an erythrocyte-lysing solution and mix. Incubate at room temperature in the darkness (the blood should be well mixed with the lysing solution). Centrifuge at 540xg for 5 minutes. The supernatant is removed with a Pasteur pipette or with a vacuum pump.
7. Resuspend and a made a final wash with 3-5 mL of PBS at 540xg for 5 min.
8. After removing the supernatant and resuspending the cell pellet, add 300 µL of PBS and adquire on the flow cytometer are recorded.
9. Analyse on a flow cytometer or store at 2- 8 °C in the dark until analysis. Samples can be run up to 24 hours after lysis.

## 6. REFERENCES


1. Zapata JM, Pulido R, Acevedo A, Sánchez-Madrid F, de Landázuri MO. Human CD45RC specificity. A novel marker for T cells at different maturation and activation stages. J Immunol. 1994 Apr 15;152(8):3852-61. PMID: 8144955.
2. Pulido, R. and Sánchez-Madrid, F. (1990), Glycosylation of CD45: carbohydrate composition and its role in acquisition of CD45RO and CD45RB T cell maturation-related antigen specificities during biosynthesis. Eur. J. Immunol., 20: 2667-2671. https://doi.org/10.1002/eji.1830201221.
3. Knapp W. Leucocyte typing IV : white cell differentiation antigens. Oxford: Oxford University Press; 1989.
4. Ostergaard HL, Trowbridge IS. Negative regulation of CD45 protein tyrosine phosphatase activity by ionomycin in T cells. Science1991 Sep 20;253(5026):1423-5.
5. Donovan JA, Koretzky GA. CD45 and the immune response. J Am Soc Nephrol1993 Oct;4(4):976-85.
6. Pulido, R., M. Cebrian, A. Acevedo, M. O. de Landazuri, and F. Sanchez-Madrid. 1988. Comparative biochemical and tissue distribution study of four distinct CD45 antigen specificities. J. Immunol. 140:3851


## 7. EXPLANATION OF SYMBOLS

| | |
|---|---|
| | Form |
| REF | Catalog reference |
| Σ | Contains sufficient for > test |
| | Quantity per test |
| | Regulatory Status |
| RUO | Research Use Only |
| [A] | Concentration |
| | Manufacturer |

## 8. MANUFACTURED BY:


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