





# Anti-Human CD34 (581)

	<b>REF</b>			<b>[A]</b>	
FITC	34F-100T	100 test	20 µL/test	0,05 mg/ml	<b>RUO</b>
PerCP-Cyanine5.5	34PP5.5-100T	100 test	5 µL/test	0,2 mg/ml	

## 1. PRODUCT DESCRIPTION

**Clone:** 581;  
**Isotype:** IgG1;  
**Tested application:** flow cytometry;  
**Immunogen:** The anti-CD34 monoclonal antibody derives from human CD34 cells;  
**Species reactivity:** Human, Cross-Reactivity: Cynomolgus;  
**Storage instruction:** store in the dark at 2-8 °C;  
**Storage buffer:** aqueous buffered solution containing protein stabilizer and 0.09% sodium azide (NaN<sub>3</sub>);  
**Recommended usage:** Immunostep's CD34, clone 581, is a monoclonal antibody intended for identification qualitative of hematopoietic progenitor cell antigen CD34 also known as gp105-120, expressed on the majority of hematopoietic stem/progenitor cells. This reagent is effective for direct immunofluorescence staining of human tissue flow cytometric analysis using 1 test for 106 cells;

**Presentation:** liquid;  
**Source:** Supernatant proceeding from an in vitro cell culture of a cell hybridoma;  
**Purification:** Affinity chromatography;  
**Other names:** Hematopoietic progenitor cell antigen CD34, Gp105-120, My10;  
**Gene ID:** 947;  
**Molecular weight:** 105 - 120 kDa.

## 2. ANTIGEN DETAILS

**Large description:** The CD34 antigen is a single-chain transmembrane glycoprotein. The antigen is associated with human hematopoietic progenitor cells and is a differentiation stage-specific leucocyte antigen.

The CD34 antigen is present on immature hematopoietic precursor cells and all hematopoietic colony-forming cells in bone marrow and blood, including unipotent and pluripotent progenitors. The CD34 antigen is present on early myeloid cells that express the CD33 antigen but lack the CD14 and CD15 antigens and on early erythroid cells that express the CD71 antigen and dimly express the CD45 antigen. The CD34 antigen is also found on capillary endothelial cells and approximately 1% of human thymocytes. Normal peripheral blood lymphocytes, monocytes, granulocytes, and platelets do not express the CD34 antigen.<sup>[1-4]</sup>

## 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 acquire 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 acquire 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. Knapp W. Leucocyte typing IV : white cell differentiation antigens. Oxford: Oxford University Press; 1989.
2. Molgaard HV, Spurr NK, Greaves MF. The hemopoietic stem cell antigen, CD34, is encoded by a gene located on chromosome 1. Leukemia 1989 Nov;3(11):773-6.
3. Hoffres HG, Lowe JA, Pedersen RO, Schmidtkte G, McDonald DF. BIRMA-K3, a new monoclonal antibody for CD34 immunophenotyping and stem and progenitor cell assay. J Hematother 1996 Jun;5(3):261-70.
4. Orfao A, Chillon MC, Bortoluci AM, Lopez-Berges MC, Garcia-Sanz R, Gonzalez M, et al. The flow cytometric pattern of CD34, CD15 and CD13 expression in acute myeloblastic leukemia is highly characteristic of the presence of PML-RARalpha gene rearrangements. Haematological 1999 May;84(5):405-12.

## 7. EXPLANATION OF SYMBOLS

	Form
<b>REF</b>	Catalog reference
	Contains sufficient for > test
	Quantity per test
	Regulatory Status
<b>RUO</b>	Research Use Only
<b>[A]</b>	Concentration
	Manufacturer

## 8. MANUFACTURED BY:

### IMMUNOSTEP S.L.

**Address:** Avda. Universidad de Coimbra, s/n  
 Cancer Research Center (C.I.C)  
 Campus de Unamuno  
 37007 Salamanca (Spain)  
**Telf./fax:** (+34) 923 294 827  
**E-mail:** [info@immunostep.com](mailto:info@immunostep.com)  
[www.immunostep.com](http://www.immunostep.com)

