# Anti-Human CD5 (CRIS-1;33-1C6)



REF	$\sum_{i=1}^{n}$	$\diamond$			5.
5PU1	l mg	lm	g/ml	1	
5F1-100T	100 test	20 µL/test	2 mg/ml		
5PE1-100T	100 test	20 µL/test	2 mg/ml	RUO	
5PPC5.51-100T	100 test	5 µL/test	0,05 mg/m		
5A1-100T	100 test	20 µL/test	2 mg/ml		
	SPUI   5F1-100T   5PE1-100T   5PPC5.51-100T   5A1-100T	REF Img   5PU1 1 mg   5F1-100T 100 test   5PE1-100T 100 test   5PPC5.51-00T 100 test   5A1-100T 100 test	REF S   5PUI 1 mg 1 mg   5FI-100T 100 test 20 µL/test   5PE1-100T 100 test 20 µL/test   5PPC5.5I-100T 100 test 5 µL/test   5AI-100T 100 test 20 µL/test	REF Img Img/ml   5PUI 1 mg 20 µL/test 2 mg/ml   5F1-100T 100 test 20 µL/test 2 mg/ml   5PE1-100T 100 test 20 µL/test 2 mg/ml   5PPC5.51-100T 100 test 5 µL/test 0,05 mg/m   5A1-100T 100 test 20 µL/test 2 mg/ml	REF Img Img/minipage Img/minipage   5PU1 1 mg 1 mg/minipage 1 mg/minipage   5F1-100T 100 test 20 µL/test 2 mg/minipage   5PE1-100T 100 test 20 µL/test 2 mg/minipage   5PPC5.51-100T 100 test 5 µL/test 0,05 mg/minipage   5A1-100T 100 test 20 µL/test 2 mg/minipage

#### 1. PRODUCT DESCRIPTION

Clone: CRIS-1 (33-1C6);

**lsotype**: lgG2a;

Tested application: flow cytometry;

Immunogen: The anti-CD5 monoclonal antibody derives from stimulated human leukocytes;

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<sub>2</sub>);

Recommended usage: Immunostep's CD5, clone 33-IC6, is a monoclonal antibody intended for the identification of T lymphocytes CD5- using flow cytometry. This reagent is effective for direct immunofluorescence staining of human tissue for flow cytometric analysis using I test for IO<sup>6</sup> cells;

Presentation: liquid;

Source: Supernatant proceeding from an in vitro cell culture of a cell hybridoma; Purification: Affinity chromatography; Other names: TI, LvI, To67, Leu-1, Lvmphocyte antigen TI/Leu-1;

Gene ID: 921;

Molecular weight: Scavenger receptor superfamily, 67 kDa.

# 2. ANTIGEN DETAILS

**Large description**: The monoclonal antibody is directed against the CD5-antigen (TI-antigen) a 67kDa transmembrane protein, which is identified as CD5 (HLDA I; WS Code T 29HLDA III; WS Code T 530), which is expressed on human T lymphocytes<sup>(0)</sup>.

The monoclonal antibody reacts with 90% of human peripheral T lymphocytes, medullary thymocytes as well as with lymphocytes of patients with chronic B-cell derived leukaemia. It is also expressed on a small subpopulation of normal B cells as a range of neoplastic B cells. The antibody does not react with, monocytes, granulocytes and platelets.<sup>[2]</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. 6.

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.

Please, refer to www.immunostep.com technical support for more information.

# 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  $^{\circ}C$  for 30 minutes or at room temperature (20-25  $^{\circ}C$ ) for 15 minutes.
- 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 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  $\mu L$  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
- 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.
- 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.
- 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  $^{\circ}\text{C}$  in the dark until analysis. Samples can be run up to 24 hours after lysis.

# REFERENCES

- Calvo J, Padilla O, Places L, Vigorito E, Vilà JM, Vilella R, Milà J, Vives J, Bowen MA, Lozano F. Relevance of individual CD5 extracellular domains on antibody recognition, glycosylation and co-mitogenic signalling. Tissue Antigens. 1999 Jul;54(1):16-26.
- Braylan RC, Orfao A, Borowitz MJ, Davis BH. Optimal number of reagents required to valuate hematolymphoid neoplasias: results of an international consensus meeting. Cytometry 2001;46:23-7.

## 7. EXPLANATION OF SYMBOLS

<b>\</b> ∎∕	Form
REF	Catalog reference
T	Contains sufficient for <n> test</n>
	Regulatory Status
$\bigcirc$	Quantity per test
RUO	Research Use Only
<b></b>	Manufacturer

#### MANUFACTURED BY: IMM



8.

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 www.immunostep.com