





Anti-Human CD11b (DCIS1/18)

	REF			[A]	
PURE	I1BPU	1 mg	1 mg/ml		
PURE	I1BPU-O1MG	100 test	1 µg/test	0,01 mg/ml	RUO
FITC	I1BF-100T	100 test	20 µL/test	2 mg/ml	

1. PRODUCT DESCRIPTION

Clone: DCIS1/18;
Isotype: IgG2a;
Tested application: flow cytometry;
Immunogen: The anti-CD11b monoclonal antibody derives from C57BL/10 splenocytes;
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 (NaN₃);
Recommended usage: Immunostep's CD11b, clone DCIS1/18, is a monoclonal antibody intended for the identification and enumeration of human leucocytes using flow cytometry. This reagent is effective for direct immunofluorescence staining of human tissue for 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: Integrin alpha-M; Ly-40; Mac-1a; Mac-1 alpha; CR3A; CR-3 alpha chain;
Gene ID: 3684;
Molecular weight: 170 kDa.

2. ANTIGEN DETAILS

Large description: The monoclonal antibody is directed against the CD11b-antigen (MO-1) also called Integrin alpha M, which is expressed on monocytes, granulocytes and NK-cells. Integrin alpha-M comprises the receptor (CR3 am chain, CD11b/CD18) for the complement component C3i.

Integrin alpha-M/beta-2 is implicated in various adhesive interactions of monocytes, macrophages and granulocytes as well as in mediating the uptake of complement-coated particles.⁽¹⁻⁴⁾

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.

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

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. Sanchez-Madrid F, Krensky AM, Ware CF, Robbins E, Strominger JL, Burakoff SJ, et al. Three distinct antigens associated with human T-lymphocyte-mediated cytotoxicity: LFA-1, LFA-2, and LFA-3. Proc Natl Acad Sci U S A 1982 Dec;79(23):7489-93.
2. Sanchez-Madrid F, Nagy JA, Robbins E, Simon P, Springer TA. A human leukocyte differentiation antigen family with distinct alpha-subunits and a common beta-subunit: the lymphocyte function-associated antigen (LFA-1), the C3bi complement receptor (OKM1/Mac-1), and the p150,95 molecule. J Exp Med 1983 Dec 1;158(6):1785-803.
3. Corbi AL, Larson RS, Kishimoto TK, Springer TA, Morton CC. Chromosomal location of the genes encoding the leukocyte adhesion receptors LFA-1, Mac-1 and p150,95. Identification of a gene cluster involved in cell adhesion. J Exp Med 1988 May 1;167(5):1597-607.
4. Springer TA, Dustin ML, Kishimoto TK, Marlin SD. The lymphocyte function-associated LFA-1, CD2, and LFA-3 molecules: cell adhesion receptors of the immune system. Annu Rev Immunol 1987;5:223-52.
5. Patarroyo M, Prieto J, Beatty PG, Clark EA, Gahmberg CG. Adhesion-mediating molecules of human monocytes. Cell Immunol 1988 May;113(2):278-89.

7. EXPLANATION OF SYMBOLS

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

8. MANUFACTURED BY:

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