# Anti-Human CD49c (VJ1/6)

| | REF | Σ | | [A] | |
|---|---|---|---|---|---|
| PURE | 49CPU | 1 mg | 1 mg/ml | 1 mg/ml | RUO |
| FITC | 49CF-100T | 100 test | 20 µL/test | 2 mg/ml | |
| PE | 49CPE-100T | 100 test | 20 µL/test | 2 mg/ml | |

## 1. PRODUCT DESCRIPTION

**Clone:** VJ1/6;
**Isotype**: IgG1;
**Tested application**: flow cytometry;
**Immunogen**: The anti-CD49c monoclonal antibody derives from TNF activated HUVEC cells;
**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 CD49c, clone TEA1/41 is a monoclonal antibody intended for the identification and enumeration of Integrin alpha-3 protein 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**: Integrin alpha-3, CD49 antigen-like family member C, FRP-2, Galactoprotein B3, GAPB3, VLA-3 subunit alpha, MSK18, VCA-2; VL3A; VLA3a;
**Gene ID:** 3673;
**Molecular weight:** 170 kDa.

## 2. ANTIGEN DETAILS

**Large description:** The monoclonal antibody is directed against the CD49c-antigen (VLA-3 alpha-chain or Integrin alpha-3/beta-1) a receptor for fibronectin, laminin, collagen, epiligrin, thrombospondin and CSPG4. Integrin alpha-3/beta-1 provides a docking site for FAP (seprase) at invadopodia plasma membranes in a collagen-dependent manner and hence may participate in the adhesion, formation of invadopodia and matrix degradation processes, promoting cell invasion. Alpha-3/beta-1 may mediate with LGALS3 the stimulation by CSPG4 of endothelial cells migration.

Monoclonal antibody VJ1/6 reacts with α 3 integrin, a transmembrane glycoprotein which non-covalently associates with integrin β 1 (CD29) to form the α3b1 (CD49c/CD29,VLA-3) complex which is proteolytically cleaved into two disulfide linked fragments of 125 kD and 30 kD. CD49c is expressed mostly on endothelial and epithelial cells (basal epidermal layers). It is not expressed on platelets and is expressed weakly on peripheral blood leukocytes. The CD49c/CD29 complex serves as an adhesive receptor for kalinin or epiligrin. This interaction may be important for thymocyte interaction with thymic epithelium. This antibody is suitable for immunohistochemical staining on acetone-fixed frozen tissue sections.[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.

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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. Wayner EA, Carter WG. Identification of multiple cell adhesion receptors for collagen and fibronectin in human fibrosarcoma cells possessing unique alpha and common beta subunits. J Cell Biol1987 Oct;105(4):1873-84.
2. Luque A, Gomez M, Puzon W, Takada Y, Sanchez-Madrid F, Cabanas C. Activated conformations of very late activation integrins detected by a group of antibodies (HUTS) specific for a novel regulatory region (355-425) of the common beta 1 chain. J Biol Chem1996 May 10;271(19):11067-75.
3. Liddington RC, Ginsberg MH. Integrin activation takes shape. J Cell Biol2002 Sep 2;158(5):833-9.
4. Mitchell K, Szekeres C, Milano V, Svenson KB, Nilsen-Hamilton M, Kreidberg JA, et al. Alpha3beta1 integrin in epidermis promotes wound angiogenesis and keratinocyte-to-endothelial-cell crosstalk through the induction of MRP3. J Cell Sci2009 Jun 1;122(Pt 11):1778-87.


## 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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