

Anti-Human CD55 (JS11)



1. PRODUCT DESCRIPTION

Clone: JS11;
Isotype: IgG1;
Tested application: flow cytometry;
Immunogen: The anti-CD55 monoclonal antibody derives from Human Peripheral Blood Mononuclear Cells and Tonsil 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 (NaN₃);
Recommended usage: Immunostep's CD55, clone JS11 is a monoclonal antibody intended for the identification and enumeration of complement decay-accelerating factor protein expressed on hematopoietic cells including erythrocytes and many non-hematopoietic cells 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: Complement decay-accelerating factor, DAF;
Gene ID: I604;
Molecular weight: 41,4 kDa.

2. ANTIGEN DETAILS

Large description: CD55 clone JS11 reacts with DAF protein a glycosylphosphatidylinositol (GPI)-anchored single chain glycoprotein also known as decay-accelerating factor.

It is expressed on hematopoietic cells including erythrocytes and many non-hematopoietic cells. This protein recognizes C4b and C3b fragments that condense with cell-surface hydroxyl or amino groups when nascent C4b and C3b are locally generated during C4 and C3 activation. Interaction of DAF with cell-associated C4b and C3b polypeptides interferes with their ability to catalyze the conversion of C2 and factor B to enzymatically active C2a and Bb and thereby prevents the formation of C4b2a and C3bBb, the amplification convertases of the complement cascade. CD55 has been reported to reduce the efficiency of NK cell lysis and induce signal transduction in T cells. CD55 has also been shown to interact with CD97 and bind to Coxsackie and Echovirus.⁽¹⁻³⁾

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 µl (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 µl (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. Nowicki B, Nowicki S. DAF as a therapeutic target for steroid hormones: implications for host-pathogen interactions. *Adv Exp Med Biol*;735:83-96.
2. Caras IW, Davitz MA, Rhee L, Weddell G, Martin DW, Jr., Nussenzweig V. Cloning of decay-accelerating factor suggests novel use of splicing to generate two proteins. *Nature*1987 Feb 5-11;325(6104):545-9.
3. Osuka F, Endo Y, Higuchi M, Suzuki H, Shio Y, Fujiu K, et al. Molecular cloning and characterization of novel splicing variants of human decay-accelerating factor. *Genomics*2006 Sep;88(3):316-22.

7. EXPLANATION OF SYMBOLS

| | |
|---|--------------------------------|
|  | Form |
|  | Catalog reference |
|  | Contains sufficient for > test |
|  | Quantity per test |
|  | Regulatory Status |
|  | Research Use Only |
|  | 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
www.immunostep.com