

Analyte Specific Reagent.

Analytical and performance characteristics are not established.

SPECIFICITY

The CD63 antigen, also known as lysosomal membrane associated glycoprotein 3 (LAMP3), is a member of the tetraspanin (TM4SF) family (1). Nearly 20 genes encode tetraspanins whose main structural characteristics is their four transmembrane domains. CD63, as other tetraspanins (CD9, CD81, CD82), has recently been reported as forming complexes with VLA-3 and phosphatidylinositol 4-kinase (2, 3), with VLA-6 (4), CD11/CD18 and tyrosine kinase (5). CD63 was first described in granules of resting platelets and on the surface membrane on activated platelets (6) The molecular weight of the platelet form of CD63 is ranging from 40 to 55 kDa (8). CD63 has been detected on the surface and in the cytoplasm of many cells (8-12)

Its surface expression is associated to lysosomal secretion: CD63, present only in azurophilic granules of non stimulated neutrophils is strongly expressed at the surface of neutrophils after activation (8, 9); this phenomenon has been also reported for basophils (10, 11).

The CLBGran/12 monoclonal antibody (mAb) reacts with most peripheral blood cells including activated platelets, lymphoid, myeloid and endothelial cells, except with red blood cells and resting T cells.

CLBGran/12 mAb has been assigned to the CD63 cluster of differentiation at the IVth International Workshop on Human Leucocyte Differentiation Antigens in Vienna, Austria, in 1989 (8) (other designations: mAb435 (8), CLB-435 (9) or CLB-CD63 (11)).

REAGENT

IOTest CD63-FITC Conjugated Antibody
PN IM1165U – 2 mL Liquid – 20 µL / test*.

Clone CLBGran/12

Isotype IgG1, mouse

Immunogen Human cytochrome B enriched cells

Hybridoma Myeloma SP2/0 Ag 1.4 x (Balb/c x AJ) spleen cells

Source Ascites fluid

Purification Ion exchange or affinity chromatography

Conjugation FITC (Fluorescein isothiocyanate) is conjugated at 15 – 25 moles of FITC per mole of Ig.

Fluorescence FITC (Green)
Excites at 468 – 509 nm
Emits at 504 – 541 nm

REAGENT CONTENTS

This reagent is provided in phosphate-buffered saline, with 0.1% sodium azide (NaN₃) as preservative, and 2.0 mg / mL bovine serum albumin (BSA).

STATEMENT OF WARNINGS

1. This reagent contains 0.1% sodium azide. Sodium azide under acid conditions yields hydrazoic acid, an extremely toxic compound. Azide compounds should be flushed with running water while being discarded. These precautions are recommended to avoid deposits in metal piping in which explosive conditions can develop. If skin or eye contact occurs, wash excessively with water.
2. Do not use antibody beyond the expiration date on the label.
3. Samples and all material coming in contact with them should be handled as if capable of transmitting infection and disposed of with proper precautions.
4. Never pipet by mouth and avoid contact of samples with skin and mucous membranes
5. Minimize exposure of reagent to light during storage or incubation.
6. Avoid microbial contamination of reagents or incorrect results might occur.
7. Use good laboratory practices when handling this reagent.

STORAGE CONDITIONS AND STABILITY

This reagent is stable up to the expiration date when stored at 2 – 8°C. Do not freeze. Minimize exposure to light.

EVIDENCE OF DETERIORATION

Any change in the physical appearance of this FITC-labeled reagent (clear, colorless to yellowish-green liquid) or any major variation in values obtained for control samples may indicate deterioration and the reagent should not be used.

REAGENT PREPARATION

No preparation is necessary. This monoclonal antibody may be used directly from the vial. Bring reagent to 18 – 25°C prior to use.

SELECTED RESEARCH REFERENCES

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2. Berditchevski, F., Tolias, K. F., Wrong, K., Carpenter, C. L., Hemler, M. E., "A novel link between integrins, transmembrane-4 superfamily proteins (CD63 and CD81), and phosphatidylinositol4-kinase", 1997, J. Biol. Chem., 272, 2595-2598.
3. Rubinstein, E., Le Naour, F., Lagaudrière-Gesbert, C., Billard, M., Conjeaud, H., Boucheix, C., "CD9, CD63, CD81, and CD82 are components of a surface tetraspan network connected to HLA-DR and VLA

integrins", 1996, Eur. J. Immunol., 26, 2657-2665.

4. Berditchevski, F., Bazzoni, G., Hemler, M. E., "Specific association of CD63 with the VLA-3 and VLA-6 Integrins", 1995, J. Biol. Chem., 270, 17784-17790.
5. Skubitz, K.M., Campbell, K.D., Lida, J., Skubitz, A.P.N., "CD63 Associates with tyrosine kinase activity and CD11/CD18, and transmits an activation signal in neutrophils", 1996, J. Immunol., 157, 3617-3626.
6. Nieuwenhuis, H.K., Van Oosterhout, J.J.G., Rozemuller, E., Van Iwaarden, F., Sixma, J.J., "Studies with a monoclonal antibody against activated platelets : evidence that a secreted 53,000-molecular weight lysosome-like granule protein is exposed on the surface of activated platelets in the circulation", 1987, Blood, 70, 838-845.
7. Azorsa, D.O., Hildreth, J.E.K., "CD63 cluster report workshop", 1995, Leucocyte Typing V, White Cell Differentiation Antigens. Schlossman, S.F., et al., Eds., Oxford University Press, 1352-1353.
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9. De Haas, M., Kerst, J.M., Van der Schoot, C.E., Calafat, J., Hack, C.E., Nuijens, J.H., Roos, D., Van Oers, R.H.J., Von Dem Borne, A.E.G.Kr., "Granulocyte colony-stimulating factor administration to healthy volunteers : analysis of the immediate activating effects on circulating neutrophils", 1994, Blood., 84, 3885-3894.
10. Mul, F.P.J., Knol, E.F., Roos, D., "An improved method for the purification of basophilic granulocytes from human blood", 1992, J. Immunol. Methods, 149, 207-214.
11. Knol, E.F., Hoogerwerf, M., Mul, FP.J., Kuijpers, T.W., Roos, D., " Analysis using the myeloid panel mAb of purified basophils obtained from human blood : changes in expression during degranulation", 1989, Leucocyte Typing IV, White Cell Differentiation Antigens. W. Knapp, et al., Eds., Oxford University Press, 1042-1043.
12. Metzelaar, M.J., Sixma, J.J., Nieuwenhuis, H.K., " A new cluster activation-dependent mAb recognizing a 53-kDa lysosome-like granule protein, expressed on the plasma membrane after activation", 1989, Leucocyte Typing IV, White Cell Differentiation Antigens. W. Knapp, et al., Eds., Oxford University Press, 1042-1043.

(*) : 20 µL is the quantity of product sufficient to stain

5 x 10⁵ cells in a standard immunofluorescence assay



IOTest[®] CD63-FITC

PN IM1165U – 2 mL Liquid – 20 µL / test* – Clone CLBGran/12

PRODUCT AVAILABILITY

IOTest CD63-FITC Conjugated Antibody
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