

**Monoclonal Mouse
Anti-Human
CD11b, C3bi Receptor/RPE**
Clone 2LPM19c
Code No. R 0841

For research use only. Not for use in diagnostic procedures.

Recommended use	Monoclonal Mouse Anti-Human CD11b, C3bi Receptor/RPE, is recommended for use in flow cytometry for identification of cells expressing CD11b.
Antigen synonyms	Integrin α L β 2, iC3b receptor, CR3.
Introduction	<p>The CD11b antigen is the 165 kDa α-chain of the Mac-1 complex. The molecule, also called CR3, functions as a cell surface receptor for the C3bi complement fragment. There is evidence that CD11b can bind fibrinogen in addition to C3bi (1). Two binding sites have now been reported, one recognizing lipopolysaccharide, and the other protein ligands containing the arg-gly-asp sequence (2). Binding of the coagulation factor X has also been suggested as an additional receptor role of CD11b (3).</p> <p>This protein belongs to a family of related heterodimeric proteins designated CD11 in the classification system for human leucocyte differentiation antigens, which shares a common β-chain with a MW of 95 kDa (CD18). Besides the C3bi receptor, this protein family contains two other members, leucocyte function associated-1 (LFA-1, CD11a) antigen and p150,95 protein (CD11c) (4).</p> <p>Each of the three CD11 α-chains are encoded by genes which cluster on the short arm of chromosome 16 between bands p11 and p13.1. The β-chain is encoded on chromosome 21 at band q22 (5).</p> <p>Each of the three members of the CD11/CD18 family appears to play a role in adhesion of leucocytes to vascular endothelium (6-9). However, the interaction between these molecules and other cell types is complex and may involve more than a single receptor (10). The three members are part of the integrins which constitute a family of related membrane receptors in cell-cell and cell-matrix interactions.</p>
Reagent provided	<p>Purified monoclonal mouse antibody conjugated with R-phycoerythrin (RPE). The conjugate is provided in liquid form in buffer containing 1% bovine serum albumin (BSA) and 15 mmol/L NaN₃, pH 7.2. Each vial contains 100 tests (10 μL of conjugate for up to 10⁶ leucocytes from normal human peripheral blood).</p> <p><u>Clone:</u> 2LPM19c. <u>Isotype:</u> IgG1, kappa. <u>Conjugate concentration mg/L:</u> See label on vial.</p>
Immunogen	Purified iC3b receptor.
Specificity	<p>Anti-CD11b, 2LPM19c, was included in the Fourth International Workshop and Conference on Human Leucocyte Differentiation Antigens, and studies by a number of laboratories confirmed its reactivity (11). It is equivalent in specificity to antibodies like Mo1 and Mac-1 (12, 13).</p> <p>Anti-CD11b, 2LPM19c, labels peripheral blood granulocytes, approximately 80% of monocytes, and a subpopulation of "null cell" peripheral lymphocytes (CD2+, CD3-) containing most of the circulating natural killer cells (12). It also labels macrophages in many tissues, although its spectrum of reactivity against these cells is narrower than that of antibodies such as Anti-CD68, clone KP1, and Anti-CD11c, clone KB90.</p>
Precautions	<ol style="list-style-type: none">1. The device is not intended for clinical use including diagnosis, prognosis, and monitoring of a disease state, and it must not be used in conjunction with patient records or treatment.2. This product contains sodium azide (NaN₃), a chemical highly toxic in pure form. At product concentrations, though not classified as hazardous, sodium azide may react with lead and copper plumbing to form highly explosive build-ups of metal azides. Upon disposal, flush with large volumes of water to prevent metal azide build-up in plumbing.3. As with any product derived from biological sources, proper handling procedures should be used.
Storage	Store in the dark at 2-8 °C. Do not use after expiration date stamped on vial. If unexpected staining is observed which cannot be explained by variations in laboratory procedures and a problem with the product is suspected, contact our Technical Services.
Staining procedure	<ol style="list-style-type: none">1. Transfer 100 μL of anticoagulated (EDTA) blood to a 12 x 75 mm polystyrene test tube.2. Add 10 μL of R 0841 and mix gently with a vortex mixer. The 10 μL is a guideline only; the optimal volume should be determined by the individual laboratory.3. The recommended negative control is a non-reactive RPE-conjugated antibody of the same isotype.4. Incubate in the dark at 4 °C for 30 minutes or at room temperature (20-25 °C) for 15-30 minutes.5. Add 100 μL of DakoCytomation Uti-Lyse™ (code Nos. S 3325 or S 3350) Reagent A to each sample and mix gently with a vortex mixer. Incubate for 10 minutes at room temperature in the dark.6. Add 1 mL of DakoCytomation Uti-Lyse™ Reagent B to each sample and mix gently with a vortex mixer. Incubate for 10 minutes at room temperature in the dark. If another lysing reagent is used in steps 5 and 6, please follow the recommendations for that reagent.








7. Centrifuge at 300 x g for 5 minutes. Gently aspirate the supernatant and discard it leaving approximately 50 µL of fluid.
8. Add 2 mL 0.01 mol/L PBS containing 2% bovine serum albumin and resuspend the cells by using a vortex mixer.
9. Repeat step 7.
10. Resuspend pellet in an appropriate fluid for flow cytometry, e.g. 0.3 mL PBS. The PBS should contain 1% paraformaldehyde (fixative) if samples are not analysed the same day.
11. 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.

Optimal conditions may vary depending on specimen and preparation method, and should be determined by each individual laboratory. It is recommended to include a suitable positive and negative control sample with each run for reagent and preparation control. Note that fluorochrome conjugates are light sensitive, and samples should be protected from light during the staining procedure and until the analysis.

References

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Explanation of symbols

 REF	Catalogue number		Keep away from sunlight (consult storage section)		Manufacturer
	Consult instructions for use		BATCH code		
	Temperature limitation		Use by		