

Monoclonal Mouse
Anti-Human CD27/FITC
Clone M-T271
Code No. F 7178

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

Recommended use	Monoclonal Mouse Anti-Human CD27/FITC, is recommended for use in flow cytometry for identification of cells expressing CD27.
Introduction	<p>CD27, a member of the nerve growth factor receptor (NGFR) superfamily, is a type I transmembrane protein consisting of a disulphide-linked 120 kDa homodimer (1-3). CD27 is expressed on mature thymocytes and on the majority of human peripheral blood T cells, on both CD4⁺ and CD8⁺ subsets. CD27 is also expressed on activated B cells and a fraction of resting natural killer (NK) cells (4, 5). Among CD4⁺ T cells, CD27 is preferentially expressed on unprimed CD4⁺CD45RA⁺CD45RO⁻ T cells, while primed CD4⁺CD45RA⁺CD45RO⁺ T cells express low levels of CD27 (2). During activation, the expression of CD27 is increased on B cells and unprimed T cells, while primed T cells have a relatively low capacity for upregulation of CD27 (3, 6). The activation is also accompanied by the appearance of a 32 kDa soluble form of CD27 (sCD27) found in supernatants of activated lymphocytes (6).</p> <p>CD27 is believed to play a role in T cell activation, since the expression is upregulated during T cell activation (1, 7).</p>
Reagent provided	<p>Purified monoclonal mouse antibody conjugated with fluorescein isothiocyanate isomer 1 (FITC). 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> M-T271. <u>Isotype:</u> IgG1, kappa. <u>Conjugate concentration mg/L:</u> See label on vial.</p>
Immunogen	Leukaemic peripheral blood T cells from a patient with chronic T cell leukaemia (T-CLL).
Specificity/reactivity	<p>Anti-CD27, M-T271, was included in the IV International Workshop and Conference on Human Leucocyte Differentiation Antigens, and studies by different laboratories confirmed its reactivity with the CD27 antigen (8). Anti-CD27, M-T271, reacts with a majority of T cells, and with a minor fraction of B cells and NK cells.</p> <p>Based on competitive binding studies, at least three different epitopes may be recognized on the CD27 molecule. Two antibodies, M-T271 and S152, recognize the CD27b epitope (9).</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 F 7178 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 FITC-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.








- 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 2 °C - 8 °C		Use by		