

## Product datasheet for **AM39018FC-N**

### LOC105369261 Mouse Monoclonal Antibody [Clone ID: MT1]

#### Product data:

Product Type:	Primary Antibodies
Clone Name:	MT1
Applications:	FC, IF
Recommended Dilution:	Clone MT1 can be applied in flow cytometry for analysis of blood and bone marrow samples or in immunohistochemistry using cytosputs, paraffin or frozen tissue sections. Flow cytometry: please see "Protocols" below. Labelled reagent is effectively formulated for direct immunofluorescent staining.
Reactivity:	Human
Host:	Mouse
Isotype:	IgG1
Clonality:	Monoclonal
Specificity:	Clone MT1, specific for CD43, produces mouse IgG1 immunoglobulins reactive with a 95-115 kD highly sialated glycoprotein. Clone MT1 antibody is used to identify B cell lines and myeloma cells. It may be used in the diagnosis of chronic lymphocytic leukemias (CLL), as an alternative to CD5. The antigen detected by MT1 is the only marker expressed on neoplasms of the very early precursor cells. In immunohistochemistry it reacts with T cells, macrophages, myeloid cells and B cells (weak). It is used for the typing of lymphomas in paraffin sections [3-5].
Formulation:	0.01 M sodium phosphate, 0.15 M NaCl, pH 7.3, 0.2% BSA, 0.09% sodium azide Label: FITC State: Liquid purified Ig fraction Label: <u>Cat. No. Label EX-max (nm) / EM-max (nm)</u> : AM39018FC-N FITC 488 / 519 AM39018RP-N 488, 532 / 578 AM39018PU-N Pure . /
Concentration:	lot specific
Purification:	Affinity chromatography
Conjugation:	FITC
Storage:	Store the antibody undiluted at 2-8°C. Fluorochrome labelled product is photosensitive and should be protected from light.
Stability:	Shelf life: one year from despatch.



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**Database Link:** [Entrez Gene 6693 Human](#)[Entrez Gene 105369261 Human P16150](#)

**Background:** CD43 antigen is expressed on all T cells, NK cells, myeloid cells and monocytes as well as on CD5 positive mature B cells. It is also present on all immature hematopoietic cells in the bone marrow [1]. The antigen is deficient in patients with Wiskott-Aldrich syndrome. A soluble form of CD43 is present in human serum [2]. CD43 may function as an adhesion molecule via interaction with CD54 although this has not been definitely established. It may also inhibit leucocyte interactions with other cells [6]. The antigen is involved in the activation of T-cells, B cells, NK cells and monocytes [5]. The membrane proximal portion of the cytoplasmic domain mediates an association with the cytoskeleton [1].

**Synonyms:** Leukocyte sialoglycoprotein, Sialophorin, Galactoglycoprotein, SPN

**Note:**

1. Conjugates with brighter fluorochromes, like PE and APC, will have a greater separation than those with dyes like FITC. When populations overlap, the percentage of positive cells using a selected marker can be affected by the choice of fluorescent label.
2. Use of monoclonal antibodies in patient treatment can interfere with antigen target recognition by this reagent. This should be taken into account when samples are analyzed from patients treated in this fashion.
3. Reagent data performance is based on EDTA-treated blood. Reagent performance can be affected by the use of other anticoagulants.

Protocol: Flow cytometry method for use with labelled (FITC, R-PE, APC, PerCP or PerCP-Cy5.5) monoclonal antibodies

1. Add 100 µl of EDTA-treated blood (i.e. approx. 10e6 leukocytes) to a 5 ml reagent tube. The content of one tube is sufficient to perform one test.
2. Add to each tube 10 µl of labelled monoclonal antibody. (Appropriate mouse Ig isotype control samples should always be included in any labelling study). Vortex the tube to ensure thorough mixing of antibody and cells.
3. Incubate the tube for 15 minutes at room temperature in the dark.
4. Add 100 µl of a lyse reagent.
5. Incubate for 10 minutes at room temperature in the dark.
6. Add 2 ml of demineralized water and incubate for 10 minutes in the dark.
7. Centrifuge the labeled cell suspension for 2 minutes at 1000 x g.
8. Remove the supernatant and resuspend the cells in 200 µl of PBS.
9. Analyze by flow cytometry within four hours (alternatively, the cells may be fixed by 0.05% of formaline in buffered saline for analysis the next day. Some antigens are readily destroyed upon fixation and this should be taken into account when using this alternative).

Flow cytometry method for use with dual and triple combinations

1. Add 100 µl of EDTA-treated blood (i.e. approx. 10e6 leukocytes) to a 5 ml reagent tube. The content of one tube is sufficient to perform one test.

**For combinations** with anti-kappa and/or anti-lambda Ig see **Application note** below.

2. Add to each tube 20  $\mu$ l of labelled monoclonal antibody combination. (Appropriate mouse Ig isotype control samples should always be included in any labelling study).
3. Vortex the tube to ensure thorough mixing of antibody and cells.
4. Incubate the tube for 15 minutes at room temperature in the dark.
5. Add 100  $\mu$ l of a lyse reagent and mix immediately.
6. Incubate for 10 minutes at room temperature in the dark.
7. Add 2 ml of demineralized water and incubate for 10 minutes in the dark.
8. Centrifuge the labelled cell suspension for 2 minutes at 1000 x g.
9. Remove the supernatant and resuspend the cells in 200  $\mu$ l of PBS.
10. Analyze by flow cytometry within four hours (alternatively, the cells may be fixed by 0.05% of formaline in buffered saline for analysis the next day. Some antigens are readily destroyed upon fixation and this should be taken into account when using this alternative).

**Application note for anti-kappa and/or anti-lambda Ig combinations**

Add 2 ml of PBS containing 0.001% (v/v) Heparin (prewarmed to 37°C) to the cell suspension Vortex, centrifuge (2 min at 300x g) and discard the supernatant.

Repeat this step twice.

Resuspend the pelleted blood cells in 100  $\mu$ l PBS, pH 7.2, containing 0.001% (v/v) Heparin.

Flow cytometry method for use with purified monoclonal antibodies

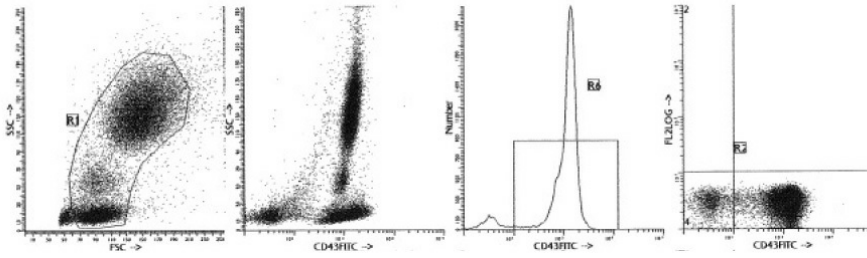
1. Add 100  $\mu$ l of EDTA-treated blood (i.e. approx. 10e6 leukocytes) to a 5 ml reagent tube. The content of one tube is sufficient to perform one test.
2. Add to each tube 10  $\mu$ l of purified monoclonal antibody. (Appropriate mouse Ig isotype control samples should always be included in any labelling study).  
Vortex the tube to ensure thorough mixing of antibody and cells.
3. Incubate the tube for 15 minutes at room temperature in the dark.
4. Wash the labelled cells by adding 2 ml of PBS containing 0.001% (v/v) Heparin, vortexing and centrifuging (2 min 1000 x g) and discard the supernatant.
5. Add 50  $\mu$ l of appropriate dilution of F(ab)2 Rabbit Anti Mouse IgG fluorescent conjugate (e.g. FITC or R-PE) in PBS containing 0.001% (v/v) Heparin to the tube. It is recommended that the tube is protected from light.
6. Mix by vortexing and incubate for 15 minutes at room temperature in the dark.
7. Add 100  $\mu$ l of a lyse reagent and mix immediately.
8. Incubate for 10 minutes at room temperature in the dark.
9. Add 2 ml of demineralized water and incubate for 10 minutes in the dark.
10. Centrifuge the labelled cell suspension for 2 minutes at 1000 x g.
11. Remove the supernatant and resuspend the cells in 200  $\mu$ l of PBS.
12. Analyze by flow cytometry within four hours (alternatively, the cells may be fixed by 0.05% of formaline in buffered saline for analysis the next day. Some antigens are readily destroyed upon fixation and this should be taken into account when using this alternative).

**Protein Families:**

Druggable Genome, ES Cell Differentiation/IPS, Transmembrane

**Protein Pathways:**

Cell adhesion molecules (CAMs)

**Product images:**

Representative data Anti-CD43, clone MT1, was analyzed by flow cytometry using a blood sample from a healthy donor. The cytogram shows direct staining with 10  $\mu$ l anti-CD43 FITC and 100  $\mu$ l of whole blood.