

Product datasheet for **AM39031PU-N**

Glycophorin A (GYPA) Mouse Monoclonal Antibody [Clone ID: NaM10-6G4]

Product data:

Product Type:	Primary Antibodies
Clone Name:	NaM10-6G4
Applications:	FC, IHC
Recommended Dilution:	CD235a antibody is used to identify erythroid cells during hematopoietic differentiation. Clone NAM10-6G4 can be applied in flow cytometry for analysis of blood and bone marrow samples or in immunohistochemistry using frozen tissue sections. The reagent is effectively formulated for direct immunofluorescent staining (see "Protocols" below).
Reactivity:	Human
Host:	Mouse
Isotype:	IgG2a
Clonality:	Monoclonal
Specificity:	Clone NAM10-6G4 produces mouse IgG2a immunoglobulins directed against the human CD235a antigen (41 kD) which is expressed as a cell surface sialoglycoprotein. This antibody may be effectively used for differentiation in acute leukemias; e.g. presence of the GpA antigen on leukemia cells indicates an early erythroid cell lineage of the tumor cells. Using the CD235a antibody, a distinction can be made between erythroleukemia and other (acute) leukemias like e.g. myeloid, lymphoid or undifferentiated leukemias [3,4]. CD235a is often used in combination with detection of H-antigen and/or anti-CD36 antibodies for additional characterization of early erythroid cells. Additional immunophenotyping of early erythroleukemias can be performed by detecting myeloid-associated antigens such as CD13, CD14, CD15, CD33 and CD34, to discriminate from lymphoid lineage-associated antigens like CD2, CD7, CD10 and CD19 [6].
Formulation:	0.01M Sodium Phosphate, 0.15M NaCl, pH 7.3 State: Aff - Purified State: Liquid purified IgG fraction Stabilizer: 0.2% BSA Preservative: 0.09% Sodium Azide Label: <u>Cat. No. Label EX-max (nm) / EM-max (nm)</u> : AM39031FC-N 488 / 519 AM39031PU-N Pure . /



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Concentration:	lot specific
Purification:	Affinity Chromatography
Conjugation:	Unconjugated
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.
Gene Name:	glycophorin A (MNS blood group)
Database Link:	Entrez Gene 2993 Human P02724
Background:	Glycophorin A (GpA) antigen (CD235a) is exclusively expressed on human erythroid cells and their progenitors and not on lymphoid or granulocytic progenitor cells and therefore is a very useful marker for detection of the erythroid cell lineage [1,2]. CD235a seems to be absent from erythroid colony forming cells in bone marrow, but is present during maturation from (pro-)erythroid blast cells to mature erythrocytes. CD235a is clinically important in the classification of leukemias.
Synonyms:	Glycophorin-A, GPA, PAS-2, Sialoglycoprotein alpha, MN sialoglycoprotein

- 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: ERYTHROCYTES - Preparation of Red Blood Cell Suspension and Immunofluorescent Staining

- Mix 2.5 ml blood with 0.75 ml Dextran solution and incubate 20 minutes at 37 °C (45° angle).
- Transfer the leukocyte-containing supernatant to another tube.
- Wash the erythrocytes with PBS (containing heparin).
- Centrifuge for 10 minutes at 2000 rpm.
- Repeat this step two times.
- Prepare an erythrocyte suspension 10% in PBS containing heparin.
- Incubate 10 µl erythrocyte suspension with 100 µl monoclonal antibody for 30 minutes at room temperature in the dark.
- Add 2 ml PBS containing heparin and centrifuge 2 min at 2000 rpm.
- Repeat this step.*
- Remove the supernatant and resuspend the cells in 200 µl PBS.

*

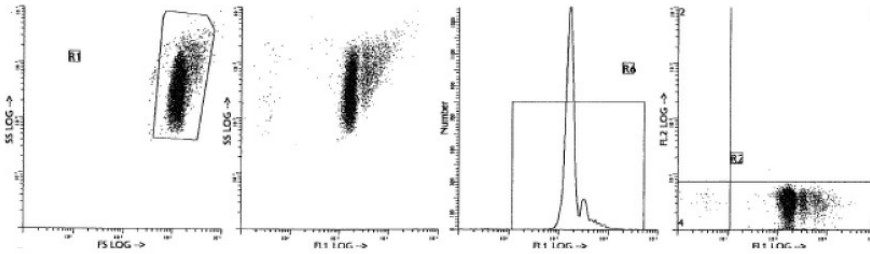
- In case of purified monoclonal antibodies:
- Add 50 µl of 1:10 dilution of F(ab)₂ Rabbit Anti Mouse IgG fluorescent conjugate in PBS containing heparin to the tube.

It is recommended that the tube is protected from light.

- Incubate for 15 minutes at room temperature in the dark.
- Add 2 ml PBS containing heparin and centrifuge 2 min at 2000 rpm.
- Remove the supernatant and resuspend the cells in 200 µl PBS.

Protein Families: ES Cell Differentiation/IPS, Transmembrane

Protein Pathways: Hematopoietic cell lineage

Product images:

Representative Data NAM10-6G4 (anti-CD235a) was analyzed by flow cytometry using a diluted blood sample (100x) from a healthy human volunteer. Direct staining was performed using 10 μ l of the FITC-conjugated monoclonal antibody preparation and 100 μ l of the diluted blood sample. N.B. No lysing solution was used.