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SZABO-SCANDIC HandelsgmbH

Quellenstraße 110, A-1100 Wien

T. +43(0)1 489 3961-0

F. +43(0)1 489 3961-7

mail@szabo-scandic.com

www.szabo-scandic.com

[linkedin.com/company/szaboscandic](https://www.linkedin.com/company/szaboscandic) 



COMBI IC Reagent: Mouse anti Myeloperoxidase-C2 (FITC) and Mouse anti Lactoferrin (PE)

nordicmubio.com/products/COMBI-IC-Reagent-Mouse-anti-Myeloperoxidase-C2-FITC-and-Mouse-anti-Lactoferrin-PE-/GIC-212-CE_slash_IVD

Catalog number: **GIC-212-CE/IVD**

Clone	8E6 and 4C5
Isotype	IgG1
Product Type	Primary Antibodies
Units	1 ml
Host	Mouse
Species Reactivity	Human
Application	Flow Cytometry

Background

Myeloperoxidase (MPO) is a glycoprotein present in the azurophil (primary) granules of myeloid cells, which appears in the myeloblast stage of myeloid cell differentiation. MPO is the most common functional protein of myeloid cells and is involved in the inflammatory response. It helps to kill microbes by breaking down peroxide in the presence of halide ions, contributing to the bactericidal function of granulocytes. The primary translation product of MPO undergoes glycosylation with production of the 89 kDa heme-free apopro-MPO form followed by incorporation of heme and conversion into the enzymatically active pro-MPO form. Subsequently, pro-MPO becomes targeted to azurophil granules where final processing occurs to produce mature dimeric MPO consisting of the 59-64 kDa MPO α -chain and the 14 kDa MPO β -chain. Lactoferrin (LF) is an iron-binding protein with bactericidal and bacteriostatic activity which is stored within the secondary granules of granulocytes. LF expression is restricted to the post-mitotic maturation compartment of the granulocytic lineage, starting from the myelocyte stage. Normal and malignant myeloblasts are LF negative. The combined staining for MPO and LF allows the distinction between mature and immature myelomonocytic cells. The MPO-C2/LF COMBI-IC reagent permits the identification and enumeration of immature and more mature myelomonocytic cell populations in normal and malignant human blood and

bone marrow using flow cytometry. Results must be put within the context of other diagnostic tests as well as the clinical history of the patient by a certified professional before final interpretation. Analyses performed with this antibody should be paralleled by positive and negative controls. If unexpected results are obtained which cannot be attributed to differences in laboratory procedures, please contact us.

Product

1 ml of FITC-conjugated anti Myeloperoxidase-C2 (clone 8E6) and PE-conjugated anti Lactoferrin (clone 4C5) in PBS pH 7.2, 1% BSA, and 0.05% NaN₃, approximately 50 tests.

Product Form: FITC and PE

Formulation: PBS pH 7.2, 1% BSA, 0.05% NaN₃

Specificity

Antibody MPO-C2 (clone 8E6) reacts with human myeloperoxidase (MPO) expressed by normal and malignant myelomonocytic cells. The LF mAb (clone 4C5) recognizes lactoferrin stored within secondary granules of postmitotic granulocyte-committed cells. In this COMBI-IC Reagent antibody 8E6 is conjugated to FITC, antibody 4C5 is conjugated to Phycoerythrin (PE). The sensitivity of MPO-C2/LF mAb is determined by staining well-defined blood samples from representative donors with serial-fold mAb dilutions to obtain a titration curve that allows relating the mAb concentration to the percentage of stained cells and geometric MFI (mean fluorescence intensity). For this purpose, a mAb-concentration range is selected to include both the saturation point (i.e. the mAb dilution expected to bind all epitopes on the target cell) and the detection threshold (i.e. the mAb dilution expected to represent the least amount of mAb needed to detect an identical percentage of cells). In practice, 50 µl of leukocytes containing 10⁷ cells/ml are stained with 20 µl mAb of various dilutions to obtain a titration curve and to identify the saturation point and detection threshold. The final concentration of the product is then adjusted to be at least 3-fold above the detection threshold. In addition and to control lot-to-lot variation, the given lot is compared and adjusted to fluorescence standards with defined intensity.

Applications

Permeabilization and Staining Procedure - In combination with our Permeabilization Kit FIX&PERM[®] (Cat. No. GAS-002) intracellular MPO-C2 and LF can be easily stained in cell suspensions. - For each sample to be analyzed add 50 µl of whole blood, bone marrow or mononuclear cell suspension in a 5 ml tube - Add 100 µl of Reagent A (Fixation Medium, stored and used at room temperature) - Incubate for 15 minutes at room temperature - Add 5 ml phosphate buffered saline and centrifuge cells for 5 minutes at 300 g - Remove supernatant and add to cell pellet 100 µl Reagent B (Permeabilization Medium) and 20 µl of the MPO-C2/LF COMBI-IC monoclonal antibody conjugate - Vortex at low speed for 1-2 seconds - Incubate for 15 minutes at room temperature - Wash cells

with phosphate buffered saline as described above - Remove supernatant and resuspend cells in sheath fluid for immediate analysis or resuspend cells in 0.5 ml 1.0 % formaldehyde and store them at 2-8°C in the dark. Analyze fixed cells within 24 hours

Storage

Nordic-MUbio monoclonal antibody reagents contain optimal concentrations of affinity-purified antibody. For stability reasons this monoclonal antibody solution contains sodium azide. These reagents should be stored at 2-8°C (DO NOT FREEZE!) and protected from prolonged exposure to light. If a slight precipitation occurs upon storage, this should be removed by centrifugation. It will not affect the performance or the concentration of the product. Stability of the reagent: Please refer to the expiry date printed onto the vial. The use of the reagent after the expiration date is not recommended.

Caution

When used for in vitro diagnostic purposes results must be put within the context of other diagnostic tests as well as the clinical history of the patient by a certified professional before final interpretation. Analyses performed with this antibody should be paralleled by positive and negative controls. If unexpected results are obtained which cannot be attributed to differences in laboratory procedures, please contact us. This product may contain hazardous ingredients. Please refer to the Safety Data Sheets (SDS) for additional information and proper handling procedures. Dispose product remainders according to local regulations. This datasheet is as accurate as reasonably achievable, but Exalpha Biologicals accepts no liability for any inaccuracies or omissions in this information.

References

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Safety Datasheet(s) for this product:

NM_Sodium Azide



Scatter characteristics of normal blood leukocyte subpopulations after immunolabeling with GIC-212.



Flow cytometric analysis of normal blood leukocyte subpopulations after immunolabeling with GIC-212.



Flow cytometric analysis of normal blood leukocyte subpopulations after immunolabeling with GIC-212.



Flow cytometric analysis of normal blood leukocyte subpopulations after immunolabeling with GIC-212.



Flow cytometric analysis of leukocyte subpopulations in normal bone marrow after immunolabeling with GIC-212.



Flow cytometric analysis of leukocyte subpopulations in leukemic bone marrow after immunolabeling with GIC-212.