Original paper DOI: https://doi.org/10.5114/ceji/210925

The significance of the classical monocyte subpopulation in chronic myelomonocytic leukemia: a single-center experience

AGNIESZKA OŻAŃSKA, JUSTYNA RYBKA, DONATA SZYMCZAK, TOMASZ WRÓBEL

Department of Hematology, Cellular Therapies, and Internal Medicine, Wroclaw Medical University, Wrocław, Poland

Abstract

Introduction: *Chronic myelomonocytic leukemia (CMML) is a myelodysplastic/myeloproliferative neoplasm. In 2022, updated diagnostic classifications for CMML were introduced by the International Consensus Classification (ICC) and World Health Organization (WHO). Monocytes are subdivided into three populations: classical (MO1), intermediate (MO2), and non-classical (MO3). One of the newly established diagnostic criteria for CMML is an increase in the MO1 fraction to ≥ 94%, as determined by multiparametric flow cytometry (MFC). This parameter has been shown to be a highly sensitive and specific marker that can rapidly and accurately differentiate CMML from other conditions.*

Material and methods: *At the Department of Hematology, Cellular Therapies, and Internal Medicine, University Clinical Hospital in Wrocław, we evaluated the distribution of monocytes and their subpopulations using MFC in 27 patients with newly diagnosed CMML, classified according to the updated WHO criteria.*

Results: *The criterion of an MO1 fraction ≥ 94% was fulfilled in 22 patients (81.5%).*

Conclusions: *Our findings are consistent with previously published data and support the utility of this method as a reliable tool for both initial screening and longitudinal monitoring of CMML.*

Key words: *chronic myelomonocytic leukemia, classical monocytes, reactive monocytosis, monocyte subsets.*

(Cent Eur J Immunol 2026; 51: 1-9)

Introduction

Chronic myelomonocytic leukemia (CMML) is one of the neoplasms within a heterogeneous group of myeloid malignancies, characterized by both myelodysplastic and myeloproliferative features [1, 2]. Over the past decade, significant advances have been made in the molecular diagnosis of myelodysplastic syndrome/myeloproliferative neoplasm (MDS/MPN), among others. This has led to changes in the classification and nomenclature of the disease. Recently, the International Consensus Classification (ICC) of myeloid neoplasms was published, updating the diagnostic criteria for CMML, among others (Table 1A) [3]. Additionally, the fifth edition of the World Health Organization (WHO) classification of hematopoietic tumors was published in 2022 (Table 1B) [3]. These classifications have been expanded to include recurrent genetic alterations, precursor states, and early stages of MDS/MPN disease, which were not included in previous versions [4].

The revised fifth edition of the WHO classification has integrated monocyte subset partitioning by multiparametric flow cytometry (MFC) as a supporting criterion for CMML diagnosis [3], while the ICC recommends searching for an abnormal monocyte profile [5].

A prerequisite for the diagnosis of CMML is an increased number of monocytes. Monocytosis has various causes; therefore, as long as CMML is not confirmed in a patient with monocytosis, a differential diagnostic process is required. The differential diagnosis in such patients should include reactive monocytosis and an increased monocyte count associated with conditions such as inflammatory processes or MPN [6].

There are three subpopulations of monocytes: classical (MO1), intermediate (MO2), and non-classical (MO3). MO1 cells strongly express the CD14 antigen and are negative for the CD16 antigen. They constitute the majority of monocytes in the blood of healthy individuals, approximately 85%. MO2 cells express CD14 and lack CD16, while MO3 cells show weak CD14 expression and positive CD16 expression. It is known that the MO1 percentage is elevated in patients with CMML. When the cut-off point for MO1 percentage in MFC is set at ≥ 94%, both the sensitivity and specificity of CMML diagnosis exceed 90%.

Correspondence: Prof. Justyna Rybka, Department of Hematology, Cellular Therapies, and Internal Medicine, Wroclaw Medical University, Wrocław, Poland, e-mail: justyna.rybka@umw.edu.pl
Submitted: 27.04.2025, Accepted: 15.09.2025

Table 1A. The ICC diagnostic criteria for chronic myelomonocytic leukemia (CMML) [3]

Monocytosis defined as monocytes ≥ 0.5 × 10^9/l and ≥ 10% of WBC
Cytopenia[a]
Blasts (including promonocytes) < 20% of nucleated cells in PB and BM
Presence of clonality Abnormal cytogenetics and/or ≥ 1 myeloid neoplasm-associated gene mutation (VAF ≥ 10%)[b]
In cases without evidence of clonality: Monocytes ≥ 1.0 × 10^9/l and ≥ 10% of WBCs with ≥ 1 of the following: – increased blasts (including promonocytes)[c] – morphologic dysplasia – abnormal immunophenotype consistent with CMML
BM examination consistent with CMML (hypercellularity due to myeloid proliferation often with increased monocytes) and lacking diagnostic features of AML, MPN or other conditions associated with monocytosis
No BCR: ABL1 fusion or genetic abnormalities consistent with M/LN-eo-TK

[a]Rare cases may show borderline or no cytopenia usually in phase disease, [b]Based on International Consensus Group Conference, Vienna, 2018 [48]; [c]Defined as blasts ≥ 5% in BM and ≥ 2% in PB
AML – acute myeloid leukemia, BM – bone marrow, CMML – chronic myelomonocytic leukemia, M/LN-eo-TK – myeloid/lymphoid neoplasms with eosinophilia and tyrosine kinase gene fusion, MPN – myeloproliferative neoplasm, PB – peripheral blood, VAF – variant allele frequency, WBCs – white blood cells

Table 1B. 2022 WHO diagnostic criteria for chronic myelomonocytic leukemia (CMML) [3]

Prerequisite criteria 1. Persistent monocytosis, defined as monocytes ≥ 0.5 × 10^9/l and ≥ 10% of WBCs
2. Blasts[a] < 20% of nucleated cells in PB and BM 3. Not meeting diagnostic criteria of CML or other MPN 4. Not meeting diagnostic criteria of M/LN-eo-TK
Supporting criteria 1. Dysplasia involving ≥ 1 myeloid lineage[b] 2. Acquired clonal cytogenetic or molecular abnormality 3. Abnormal partitioning of PB monocyte subsets[c]
Diagnostic requirements A diagnosis of CMML is made if all prerequisite criteria are present together with: – ≥ 1 supporting criterion, if monocytosis is ≥ 1 × 10^9/l – both supporting criteria #1 and #2, if monocytosis is 0.5-1.0 × 10^9/l
Subtyping criteria – Myelodysplastic CMML: WBCs < 13 × 10^9/l – Myeloproliferative CMML: WBCs ≥ 13 × 10^9/l
Subgrouping criteria (based on percentage of blasts and promonocytes) – CMML-1: < 5% in PB < 10% in BM – CMML-2: 5-19% in PB and 10-19% in BM

[a] Blast count includes myeloblasts, monoblasts and promonocytes; [b] Dysplasia should be present in ≥ 10% of cells of a hematopoietic lineage in the BM; [c] Based on detection of > 94% classical monocytes in the absence of known active autoimmune disease and/or systemic inflammatory syndromes
BM – bone marrow, CML – chronic myeloproliferative neoplasm, MPN – myeloproliferative neoplasm, PB – peripheral blood, WBCs – white blood cells

Furthermore, an MO3 fraction of < 1.13% is also useful in distinguishing CMML from other diseases [7-9]. This pattern is independent of CMML subtype, karyotype, and mutational factors. Additionally, MFC can be used to assess response to treatment.

False-negative results can occur in the presence of concurrent inflammatory conditions, including autoimmune diseases. Conversely, false-positive results may arise due to the presence of other myeloid malignancies, such as atypical chronic myeloid leukemia (aCML) [10].

The ICC has proposed revising the diagnostic criteria for CMML by lowering the threshold for monocytosis in peripheral blood (PB), adding cytopenias as a new diagnostic requirement, and emphasizing the role of genetic abnormalities in assessing disease clonality [5].

Both the 2022 WHO classification and the ICC recognize the importance of phenotypic testing in distinguishing CMML from other causes of monocytosis. Thus, the diagnosis of CMML must always be based on a combination of clinicopathological, phenotypic, and molecular data [3, 5]. Currently, there are two updated yet divergent CMML classifications, each based on partially different diagnostic criteria [11]. The differences include requirements for bone marrow (BM) characteristics, MFC results, and variant allele frequency (VAF) thresholds used to define clonality based on mutations [3, 5].

Material and methods

A retrospective analysis was conducted on patients with newly diagnosed, previously untreated CMML at the Department of Hematology, Cellular Therapies, and Internal Medicine, University Clinical Hospital in Wrocław. Peripheral blood and BM samples were collected in EDTA tubes after obtaining written informed consent, in accordance with the Declaration of Helsinki. The aim of the study was to assess the fulfillment of the WHO 2022 classification criteria for CMML, in particular the MO1 ≥ 94% threshold, among newly diagnosed patients.

A total of 27 patients diagnosed between January 2022 and June 2025 were included, comprising 7 women and 20 men. The median age was 74 years (range: 61-87 years).

Patients were evaluated based on the new 2022 WHO criteria (we did not assess patients according to the ICC classification). Monocytosis greater than 0.7 × 10^9/l was observed in their PB, with monocytes accounting for ≥ 10% of white blood cells (WBC). Monocytosis was accompanied by cytopenias and dysplasia, along with clonal genetic abnormalities. Acute myeloid leukemia (AML), MDS, CML with p190 fusion, and myeloid/lymphoid neoplasms with eosinophilia and tyrosine kinase gene fusions (M/LN-Eo-TK) were excluded. Among the various classification criteria for the disease, this study primarily focused

Table 2. Comparison of monocyte-related parameters between study group and control group

Variable	Study group (n = 27)	Control group (n = 10)	MD (95% CI)	p
MO1 in PB (%)	96.10 (94.20-97.55)	91.00 (88.90-92.80)	5.10 (2.90-7.50)	0.001
MO2 in PB (%)	2.90 (1.65-4.45)	5.60 (4.48-6.60)	–2.70 (–4.20–-1.10)	0.007
MO3 in PB (%)	0.50 (0.10-1.35)	3.20 (2.58-5.07)	–2.70 (–4.10–-1.80)	< 0.001
Monocytes in PB (%)	24.78 ±10.37	6.07 ±1.02	18.71 (14.56-22.85)	< 0.001
PB monocytosis (×10³/µl)*	2.68 (1.57-3.46)	0.45 (0.38-0.56)	2.23 (1.11-2.91)	< 0.001

MD – mean or median difference, CI – confidence interval, PB – peripheral blood. Data presented as mean± standard deviation in case of monocytes in PB or median (interquartile range) in case of other variables, depending on distribution normality. Comparisons made with Welch's t test (monocytes in PB) or Mann-Whitney U test (other variables)

** PB monocytosis available for n = 25 patients*

on the phenotypic analysis of monocyte subpopulations. Patients were examined not only for the percentage distribution of monocyte subpopulations in PB using MFC, but also to determine how many met the cut-off criterion of ≥ 94% for MO1 in CMML. Additionally, we assessed how many patients had MO3 < 1.13%. The control group consisted of 10 healthy individuals. The median age was comparable to that of the study group (Table 2).

Mouse anti-human monoclonal antibodies, all purchased from Becton Dickinson and Company (BD), San Jose, CA, were used for analysis: CD64, CD16, CD2, CD56, CD123, CD14, HLA-DR, and CD45. For MFC analysis, 4 ml of blood was collected in EDTA tubes (BD). Whole PB (150 µl) nucleated cells were surface-stained with the following fluorescence-conjugated mouse anti-human monoclonal antibodies in a single eight-color tube: CD64 FITC, CD16 PE, CD2 PerCP-Cy5.5, CD56 PC-7, CD123 APC, CD14 APC-H7, HLA-DR V450, and CD45 V500 with procedure lyse/wash. Lysing solution from BD was diluted 10-fold and used for lysing. The evaluation of nucleated cells was carried out using an eight-color FACS Canto II flow cytometer BD. In each test tube, as many cells as possible were collected, with an average of 200,000 cells per sample, resulting in an average of 50,000 monocytes with the $CD14^{+}CD16^{-}$ phenotype. The data were analyzed using BD FACSDiva software v8.0 (Fig. 1). Cell staining and sample acquisition took place immediately after the sample was delivered to the flow cytometry laboratory, i.e. within 4 hours of collection.

Statistical calculations were performed in R software (version R 4.4.2). Categorical parameters were presented with n (%). Numerical parameters were presented with mean ±standard deviation (SD) or median (interquartile range – IQR). Normality was verified with the Shapiro-Wilk test as well as skewness and kurtosis. Comparisons were made using Student's t test, Welch's t test and the Mann-Whitney U test, as appropriate. The mean/median difference (MD) with 95% confidence interval (CI) was calculated for all comparisons. Correlation analysis was conducted using the Spearman correlation method due to non-normal distribution of some variables. All statistical tests assumed significance when $p < 0.05$ (Table 3).

Results

All monocyte-related parameters demonstrated a significant difference between patients with CMML and the control group. MO1 in PB was higher among patients with CMML compared to the control group, MD = 5.10, 95% CI: 2.90 to 7.50, $p = 0.001$. MO2 in PB was lower among patients with CMML compared to the control group, MD = –2.70, 95% CI: –4.20 to –1.10, $p = 0.007$. MO3 in PB was lower among patients with CMML compared to the control group, MD = –2.70, 95% CI: –4.10 to –1.80, $p < 0.001$. The proportion of monocytes in PB was higher among patients with CMML compared to the control group, MD = 18.71, 95% CI: 14.56 to 22.85, $p < 0.001$. PB monocytosis was higher among patients with CMML compared to the control group, MD = 2.23, 95% CI: 1.11 to 2.91, $p < 0.001$ (Fig. 2).

A total of 22 patients (81.5%) fulfilled the criterion of MO1 > 94% in PB. In 19 patients (70.0%), MO3 < 1.13% was observed. Concurrent fulfillment of both conditions was noted in 18 patients (66.7%) (Table 2).

The first set of figures presents histograms, shown only for the study group, illustrating the distribution of all continuous parameters within this cohort (Fig. 3). The second figure is a box-and-whisker plot (boxplot), which provides a clear comparison of the values and distributions of the analyzed parameters between groups. Comparisons are shown only for those parameters for which data were available in both groups.

Discussion

Multiparametric flow cytometry is valuable for confirming the number of monocytes and blast cells, as well as identifying different monocyte populations. Analyzing the percentage of PB monocyte subpopulations using MFC has been proposed as a quick and effective method to differentiate CMML from reactive monocytosis, emphasizing an increase in the MO1 fraction above 94% and a decrease in the percentage of MO3. Our experience with routine MFC testing on PB samples from newly diagnosed CMML patients is similar to findings reported in the literature.

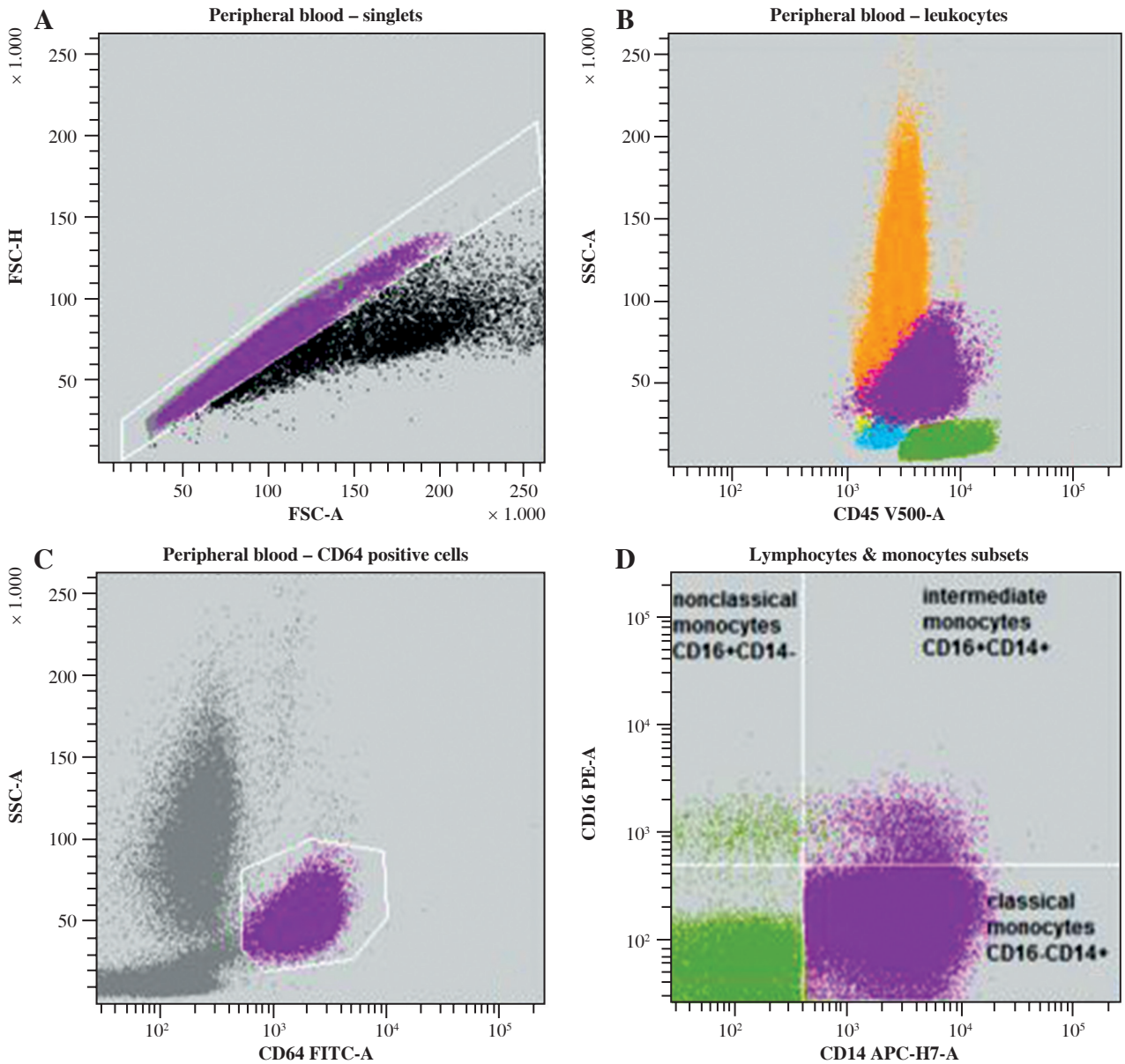

Fig. 1. Example of cytometric dot plots from a patient diagnosed with CMML and a patient with non-CMML monocytosis. Summary of the gating strategy with key dot plots – comparison: **A-D**) Patient with CMML, **A1-D1**) Patient with non-CMML monocytosis. **A, A1**) Discrimination of doublets (FSC-A vs. FSC-H); **B, B1**) Leukocyte subsets (CD45 vs. SSC-A): green – lymphocytes, blue – basophils, yellow – dendritic cells, purple – monocytes, orange – granulocytes; **C, C1**) Monocytes ($CD64^{bright+}$ cells) (CD64 vs. SSC-A); **D, D1**) Monocyte subsets (CD14 vs.CD16). **D**) The patient was diagnosed with CMML, with monocytes in peripheral blood accounting for 33.2%. The monocyte subpopulations were: classical monocytes 94.7%, intermediate monocytes 4.9%, and non-classical. **D1**) The patient with reactive monocytosis, with monocytes in peripheral blood accounting for 21.0%. The monocyte subpopulations were: classical monocytes 89.7%, intermediate monocytes 8.8%, and non-classical monocytes 1.5%. Dendritic cells should be excluded from the analysis before gating monocytes

Researchers from France [7] developed an MFC assay to distinguish MO1, MO2, and MO3 subgroups in PB mononuclear cells using a cohort of 175 patients with CMML. Compared to 307 healthy donors and patients with reactive monocytosis or other hematologic malignancies, patients with CMML showed a characteristic increase in the MO1 fraction. Their research indicates that an increase in MO1 to ≥ 94% of the total monocyte count is a highly sensitive and specific diagnostic marker that quickly and accurately distinguishes CMML from other conditions. This study,

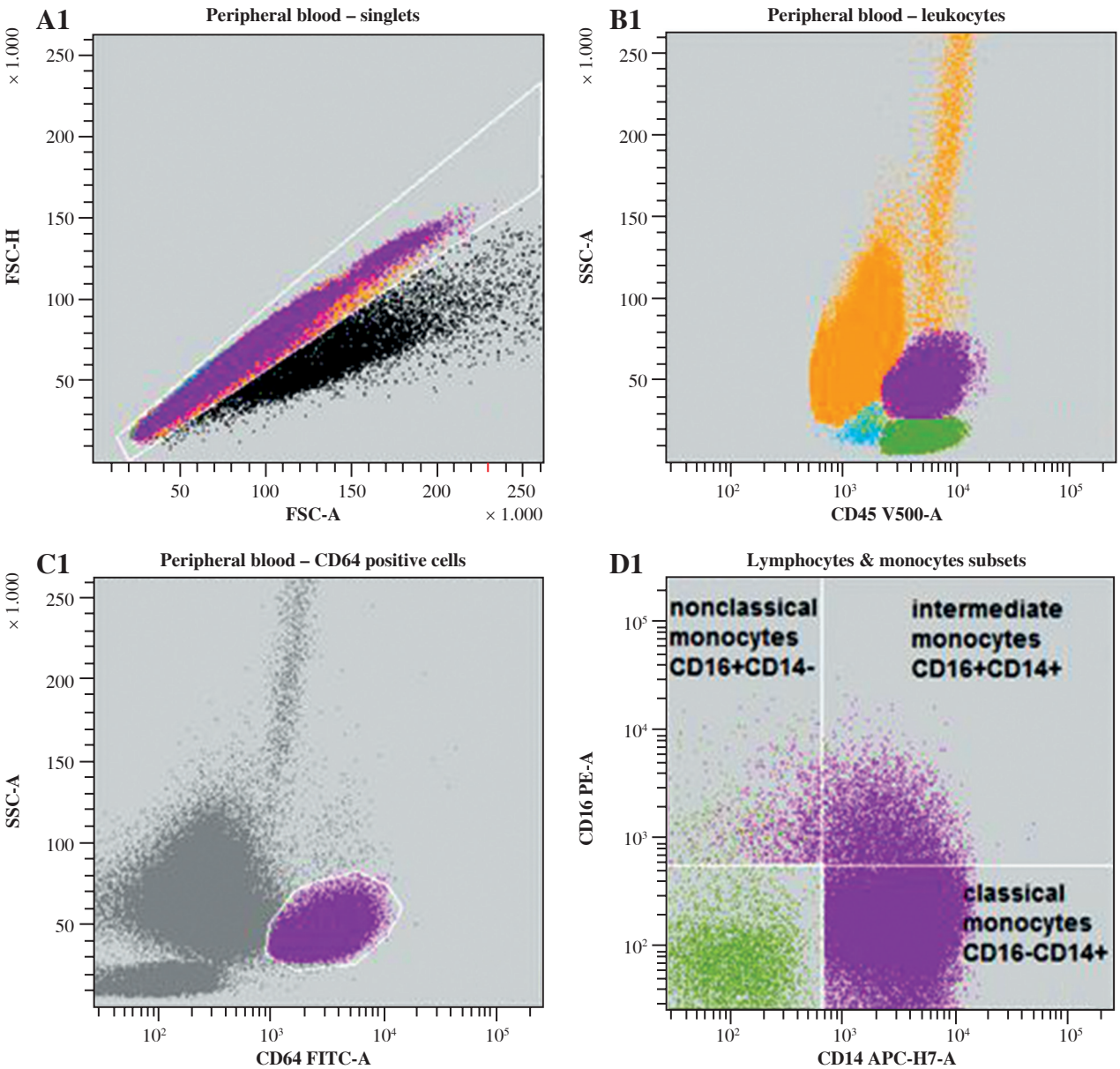

Fig. 1. Cont.

conducted on a large cohort of patients, laid the foundation for the subsequent modification of the diagnostic classification of CMML. Based on its findings, the criterion of MO1 > 94% was explicitly added as a supportive (non-mandatory) criterion for the diagnosis of CMML. Our results differ slightly, mainly due to the small group of patients originating from a single clinical center.

A group of scientists from South Korea [12] assessed PB monocyte subsets in 50 patients with CMML, reactive monocytosis, and healthy controls using MFC. They observed false-negative results in CMML patients with concurrent inflammation due to increased MO2, suggesting that inflammation was present at the time of diagnosis. We observed a similar pattern in our patients with MO1 < 94% and elevated MO2, but we did not detect active infection in them at the time of diagnosis. We evaluated only patients with *de novo* CMML, and in laboratory tests of our patients, we did not detect the simultaneous presence of other causes of monocytosis. In the remaining patients, the decrease in MO1 could potentially be related to an active infection, which may have distorted the outcome. However, in our patients, this was not confirmed by blood tests, imaging studies, microbiological tests, or clinical presentation.

The monocyte test presents challenges, including both false-positive and false-negative results. Certain situations may cause a relative accumulation of MO1, resulting in

Table 3. Study group characteristics

Variable	Mean ±SD or *n* (%)	Median (IQR)	Range
Number	27 (100.0)	–	–
Sex			
Female	7 (25.9)	–	–
Male	20 (74.1)	–	–
Age (years)	74.74 ±6.71	74.00 (70.00-80.00)	61.00-87.00
MO1 in PB (%)	94.94 ±4.45	96.10 (94.20-97.55)	81.00-99.60
MO2 in PB (%)	3.78 ±3.40	2.90 (1.65-4.45)	0.30-14.00
MO3 in PB (%)	0.94 ±1.22	0.50 (0.10-1.35)	0.00-5.00
Blasts in PB (%)*	0.48 ±0.68	0.10 (0.06-0.72)	0.00-2.60
Monocytes in PB (%)	24.78 ±10.37	22.40 (17.85-30.30)	10.00-55.30
MO1 in PB ≥ 94%	22 (81.5)	–	–
MO3 in PB < 1.13%	19 (70.0)	–	–
MO1 in PB ≥ 94% and MO3 in PB < 1.13%	18 (66.66)	–	–
PB monocytosis ($\times 10^3/\mu l$)*	3.85 ±4.66	2.68 (1.57-3.46)	0.70-19.17

SD – standard deviation, IQR – interquartile range, PB – peripheral blood
** Blasts in PB available for n = 24 patients, PB monocytosis available for n = 25 patients*

false-positive findings. A common cause of such accumulation is recovery from bone marrow aplasia. It has been shown that MO1 appear first in the marrow, followed by the sequential maturation of MO2 and then MO3 [13]. Glucocorticoid [14, 15] therapy can also lead to depletion of MO2 monocytes, resulting in a relative accumulation of MO1 and mimicking an MFC profile characteristic of CMML.

In false-negative cases, some patients with genuine CMML may exhibit an MO1 percentage below the 94% threshold. An inflammatory state may occur in 16-20% of CMML cases [16, 17]. In inflammatory conditions, a decrease in the MO3 population is observed alongside an increase in the MO2 population, leading to a concurrent reduction in the relative MO1 percentage below the typical 94% threshold, thus preventing the recognition of CMML. CMML patients in inflammatory conditions show the "bulbous" profile with an increased MO2 population [17].

The disappearance of the MO3 population is considered a hallmark of CMML. It has been confirmed that slan, a known MO3 marker, is expressed by approximately half of the MO3 population [18]. The study by Tarfi *et al*. showed that 55 CMML patients demonstrated a relative decrease in slan-positive MO3 percentage to below 1.7%. Notably, the most significant decrease was observed in seven patients exhibiting a "bulbous" profile in MFC [18]. The 1.7% threshold was established to achieve 100% sensitivity, ensuring the capture of all CMML diagnoses, particularly in cases with an inflammatory profile in MFC. A two-step algorithm has been proposed. First, the MO1 subset should be quantified; when the MO1 percentage is below 94% and only if the MFC profile displays a characteristic "bulbous" aspect, the slan-positive MO3 fraction should then be assessed. A percentage below 1.7% indicates CMML associated with an inflammatory state. Genuine reactive monocytosis may display a slan-positive MO3 fraction below 1.7%. It is recommended to add the slan antibody to the antibody panel to minimize false-negative results due to inflammation in CMML patients [19]. Unfortunately, our study is retrospective, and we did not have access to the slan antibody at the time, so it was not assessed.

An Australian study validated flow cytometry monocyte subset partitioning for CMML diagnosis. Cut-offs of > 94% classical and < 1.13% non-classical monocytes differentiated CMML from other causes of monocytosis, with sensitivities of 73-82% and specificities of 83-89% [20].

Liu *et al*. [21] analyzed 56 PB and 69 BM samples using a new gating strategy. The PB cohort included CMML, non-MN (patients without myeloid neoplasms) and non-CMML-MN (other myeloid neoplasms, e.g. MDS, AML). The BM cohort included the same groups. MO1 > 94% in blood distinguished CMML with 90% sensitivity and 88.9% specificity, while MO3 < 1.24% in marrow showed 96% sensitivity and 79.5% specificity.

Our experience is similar to findings reported in the literature. Potential reasons for lower test performance compared to its use in larger institutions may include variations in gating strategy or antibodies used. However, the main limitation of our study was the small sample size. Not all patients underwent molecular testing, or cytogenetic analysis. The ability to analyze only a limited number of markers simultaneously in MFC is another limitation. Monocyte subpopulations in our study were analyzed exclusively on monocyte cells, with lymphocytes not being considered. However, lymphocytes are shown in the dot plot presented in the study. This

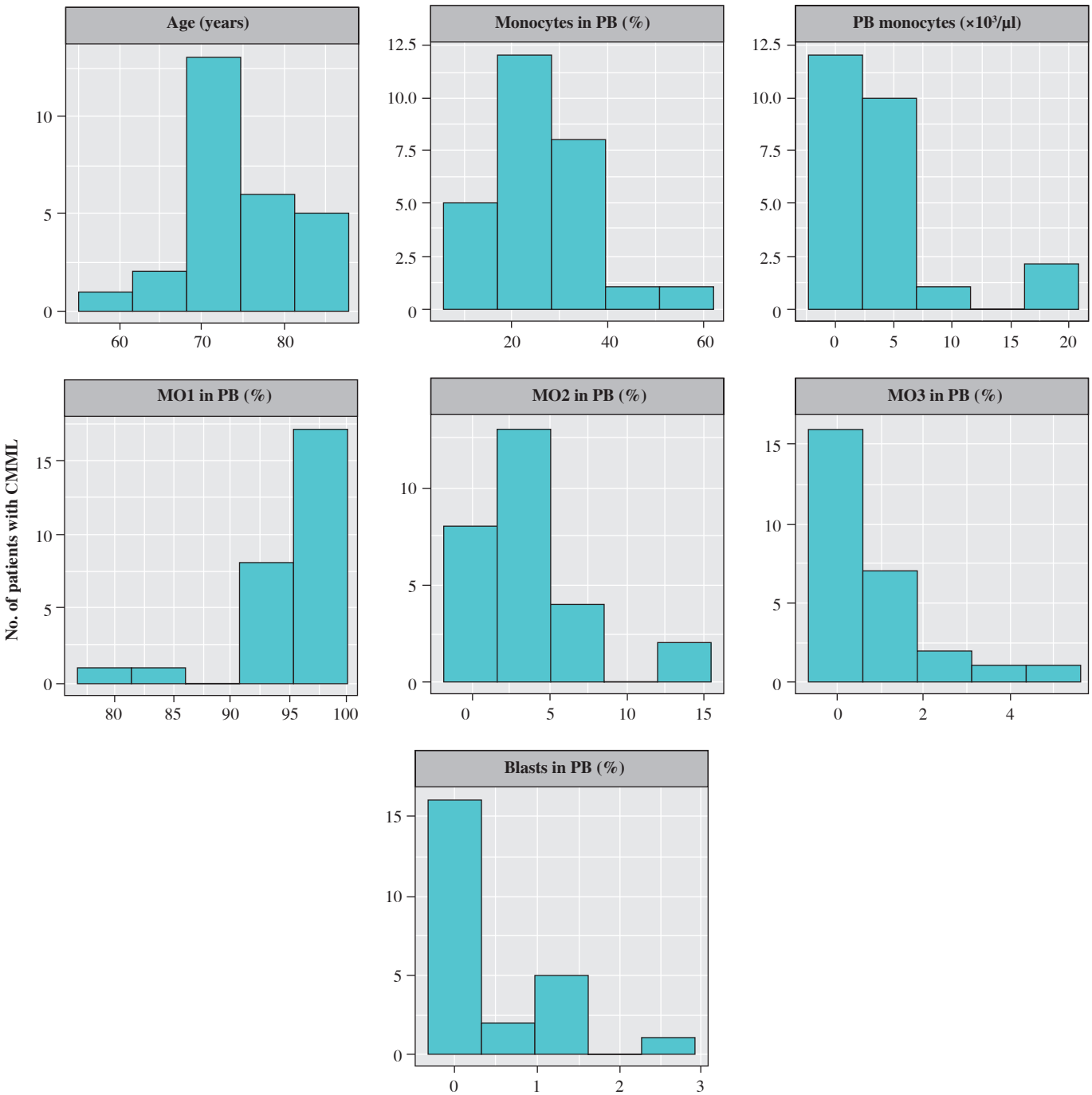

Fig. 2. Histograms of analyzed parameters, study group

is because there is often an issue with gating CD16-positive monocytes, and NK cells physiologically express CD16 on their surface, serving as a biological control for this antigen. Additionally, in the method used, monocytes were gated exclusively on strong CD64 (CD64bright^{+}). CD64 can be very dim in MO3 [22], so this gating strategy could potentially miss MO3 and lead to an overestimation of MO1. However, this is how our 8-color panel was designed, and there was no room for additional antibodies.

The study primarily focused on meeting the criterion of monocyte subpopulation phenotypes. The study findings do not influence current recommendations or therapeutic

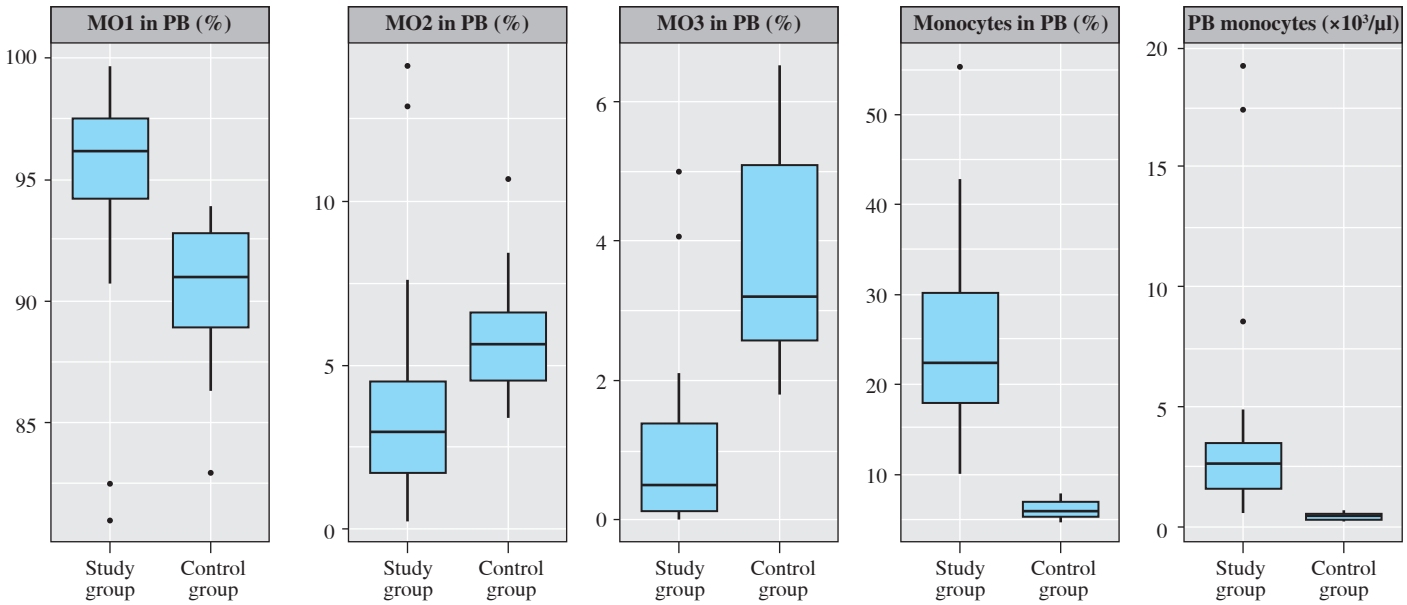

Fig. 3. Boxplots presenting distribution of monocyte-related parameters in study group and control group

strategies. Conducting research with a larger sample size might yield results more consistent with those reported in the literature. Further research is needed, potentially focusing on evaluating more diagnostic aspects of CMML, such as blast count or the presence of specific genetic mutations. Assessing our patients according to the ICC classification as well might have led to the identification of more CMML cases. Unlike the WHO classification, the ICC does not specify an exact percentage distribution of monocyte subpopulations, which was a key focus of our study, and therefore we did not take it into account.

Conclusions

In conclusion, our experience with routine MFC testing of PB samples from newly diagnosed CMML patients, according to WHO criteria, is largely consistent with previously reported findings. Potential reasons for lower test performance compared with larger institutions may include variations in gating strategies or antibody panels. To improve accuracy and reliability, a larger sample size would be necessary. Nonetheless, we believe that PB samples can be effectively used for CMML diagnosis using MFC. Monocyte subset analysis remains a valuable tool for both screening and disease monitoring. Our study was not designed to propose changes to the standardization of monocyte subset analysis protocols.

Funding

This study was supported by the grant of Wroclaw Medical University (Grant No.: SUBZ.C140.25.006).

Disclosures

The study was approved by the Bioethics Committee of the Wroclaw Medical University (Approval No. KB23/2025).

The authors declare no conflict of interest.

References

1. Patnaik MM, Lasho T (2020): Evidence-based minireview: Myelodysplastic syndrome/myeloproliferative neoplasm overlap syndromes: A focused review. Hematology 2020: 460-464.
2. Prakash S, Arber DA, Bueso-Ramos C, et al. (2022): Advances in myelodysplastic/myeloproliferative neoplasms. Virchows Arch 482: 69-83.
3. Khoury JD, Solary E, Abla O, et al. (2022): The 5th edition of the World Health Organization Classification of Haematolymphoid Tumours: Myeloid and histiocytic/dendritic neoplasms. Leukemia 36: 1703-1719.
4. Pizzi M, Gurrieri C, Orazi A (2023): What's new in the classification, diagnosis and therapy of myeloid leukemias. Hemato 4: 112-134.
5. Arber DA, Orazi A, Hasserjian RP, et al. (2022): International Consensus Classification of Myeloid Neoplasms and Acute Leukemias: Integrating morphologic, clinical, and genomic data. Blood 140: 1200-1228.
6. Ziegler-Heitbrock L, Ancuta P, Crowe S, et al. (2010): Nomenclature of monocytes and dendritic cells in blood. Blood 116: e74-80.
7. Selimoglu-Buet D, Wagner-Ballon O, Saada V, et al. (2015): Characteristic repartition of monocyte subsets as a diagnostic signature of chronic myelomonocytic leukemia. Blood 125: 3618-3626.
8. Talati C, Zhang L, Shaheen G, et al. (2017): Monocyte subset analysis accurately distinguishes CMML from MDS and

is associated with a favorable MDS prognosis. Blood 129: 1881-1883.
9. Tarfi S, Harrivel V, Dumezy F, et al. (2017): Multicentric validation of the "monocyte assay" for chronic myelomonocytic leukemia diagnosis by flow cytometry. Blood (ASH Annual Meeting Abstracts) 130 (Suppl): 4273.
10. Pophali PA, Timm MM, Mangaonkar AA, et al. (2019): Practical limitations of monocyte subset repartitioning by multiparametric flow cytometry in chronic myelomonocytic leukemia. Blood Cancer J 9: 65.
11. Patnaik MM, Zeidan AM, Padron E, et al. (2022): Differences in classification schemata for myelodysplastic/myeloproliferative overlap neoplasms. Leukemia 36: 2934-2938.
12. Hwang SM, Ahn H, Jeon S, et al. (2021): Monocyte subsets to differentiate chronic myelomonocytic leukemia from reactive monocytosis. J Clin Lab Anal 35: e23576.
13. Patel AA, Zhang Y, Fullerton JN, et al. (2017): The fate and lifespan of human monocyte subsets in steady state and systemic inflammation. J Exp Med 214: 1913-1923.
14. Fingerle-Rowson G, Angstwurm M, Andreesen R, Ziegler-Heitbrock HW (1998): Selective depletion of CD14+ CD16+ monocytes by glucocorticoid therapy. Clin Exp Immunol 112: 501-506.
15. Heimbeck I, Hofer TPJ, Eder C, et al. (2010): Standardized single-platform assay for human monocyte subpopulations: Lower CD14+CD16++ monocytes in females. Cytometry A 77: 823-830.
16. Zahid MF, Barraco D, Lasho TL, et al. (2017): Spectrum of autoimmune diseases and systemic inflammatory syndromes in patients with chronic myelomonocytic leukemia. Leuk Lymphoma 58: 1488-1493.
17. Selimoglu-Buet D, Badaoui B, Benayoun E, et al. (2017): Accumulation of classical monocytes defines a subgroup of MDS that frequently evolves into CMML. Blood 130: 832-835.
18. Tarfi S, Badaoui B, Freynet N, et al. (2020): Disappearance of slan-positive non-classical monocytes for diagnosis of chronic myelomonocytic leukemia with an associated inflammatory state. Haematologica 105: e147-e152.
19. Tarfi S, Kern W, Goulas E, et al. (2024): Technical, gating and interpretation recommendations for the partitioning of circulating monocyte subsets assessed by flow cytometry. Cytometry B Clin Cytom 106: 203-215.
20. Barge L, Gooch M, Hendle M, Simleit E (2023): Real world implementation of flow cytometric monocyte subset partitioning for distinguishing chronic myelomonocytic leukaemia from other causes of monocytosis. Pathology 55: 827-834.
21. Liu Y, Tariq H, Fu L, et al. (2025): Usefulness of flow cytometry monocyte partitioning in the diagnosis of chronic myelomonocytic leukemia in a real-world setting. Cancers (Basel) 17: 1229.
22. Jurado R, Huguet M, Xicoy B, et al. (2023): Optimization of monocyte gating to quantify monocyte subsets for the diagnosis of chronic myelomonocytic leukemia. Cytometry B Clin Cytom 104: 319-330.