Brief Communication
Diagnostic Hematology

Ann Lab Med 2018;38:481-483
https://doi.org/10.3343/alm.2018.38.5.481
ISSN 2234-3806 • eISSN 2234-3814

ANNALS OF LABORATORY MEDICINE

The 2016 WHO versus 2008 WHO Criteria for the Diagnosis of Chronic Myelomonocytic Leukemia

Yeonsook Moon, M.D.[1], Mi Hyang Kim, M.D.[2], Hye Ryoun Kim, M.D.[3], Jeong-Yeal Ahn, M.D.[4], Jungwon Huh, M.D.[5], Ji Young Huh, M.D.[6], Jae Ho Han, M.D.[7], Joon Seong Park, M.D.[8], and Sung Ran Cho, M.D.[9]

Department of Laboratory Medicine[1], Inha University School of Medicine, Incheon; Department of Laboratory Medicine[2], Kosin University College of Medicine, Busan; Department of Laboratory Medicine[3], Chung-Ang University College of Medicine, Seoul; Department of Laboratory Medicine[4], Gachon University Gil Hospital, Incheon; Department of Laboratory Medicine[5], College of Medicine, Ewha Womans University, Seoul; Department of Laboratory Medicine[6], CHA Bundang Medical Center, CHA University, Seongnam; Departments of Pathology[7], Hematology-Oncology[8], and Laboratory Medicine[9], Ajou University School of Medicine, Suwon, Korea

The 2016 WHO diagnostic criteria for chronic myelomonocytic leukemia (CMML) require both absolute and relative monocytosis ($\geq 1 \times 10^9$/L and $\geq 10\%$ of white blood cell counts) in peripheral blood. Moreover, myeloproliferative neoplasm (MPN) features in bone marrow and/or MPN-associated mutations tend to support MPN with monocytosis rather than CMML. We assessed the impact of the 2016 WHO criteria on CMML diagnosis, compared with the 2008 WHO criteria, through a retrospective review of the medical records of 38 CMML patients diagnosed according to the 2008 WHO classification. Application of the 2016 WHO criteria resulted in the exclusion of three (8%) patients who did not fulfill the relative monocytosis criterion and eight (21%) patients with an MPN-associated mutation. These 11 patients formed the 2016 WHO others group; the remaining 27 formed the 2016 WHO CMML group. The significant difference in the platelet count and monocyte percentage between the two groups indicated that the 2016 WHO criteria lead to a more homogenous and improved definition of CMML compared with the 2008 WHO criteria, which may have led to over-diagnosis of CMML. More widespread use of molecular tests and more sophisticated clinical and morphological evaluations are necessary to diagnose CMML accurately.

Key Words: Chronic myelomonocytic leukemia, WHO classification, Monocytosis

Received: October 29, 2017
Revision received: December 12, 2017
Accepted: April 27, 2018

Corresponding author: Sung Ran Cho
https://orcid.org/0000-0001-8898-3301
Department of Laboratory Medicine,
Ajou University School of Medicine,
164 World cup-ro, Yeongtong-gu,
Suwon 16499, Korea
Tel: +82-31-219-5780
Fax: +82-31-219-5778
E-mail: sungran@aumc.ac.kr

© Korean Society for Laboratory Medicine
This is an Open Access article distributed under the terms of the Creative Commons Attribution Non-Commercial License (http://creativecommons.org/licenses/by-nc/4.0) which permits unrestricted non-commercial use, distribution, and reproduction in any medium, provided the original work is properly cited.

Chronic myelomonocytic leukemia (CMML) is a clinically heterogeneous disorder with poor prognosis [1]. CMML, once classified as a MDS according to the French-American-British classification, is now recognized by the WHO classification as an overlap MDS/myeloproliferative neoplasm (MPN) [1]. The diagnosis of CMML can be difficult because it requires a combination of morphological, histopathological, and cytogenetic approaches [2]. MPN with monocytosis can simulate CMML in the absence of a previous history of MPN [3].

The 2016 WHO classification has resulted in several changes in the diagnosis and classification of CMML [4]. First, in contrast with the 2008 WHO classification [5], the diagnosis of CMML requires both the presence of absolute monocytosis ($\geq 1 \times 10^9$/L) and relative monocytosis ($\geq 10\%$) in peripheral blood (PB). Second, the presence of MPN features in the bone marrow (BM) and/or of MPN-associated mutations such as *JAK2, CALR*, and *MPL*, tend to support a diagnosis of MPN with monocytosis rather than CMML. Third, a further subdivision has split CMML-1 into CMML-0 and CMML-1. Fourth, CMML is divided into two subtypes—the proliferative type and the dysplastic type—based on white blood cell (WBC) counts.

The present study aimed to clarify the clinical and diagnostic

significance of the 2016 WHO classification for CMML and to elucidate the current utilization status of molecular genetic profiles including MPN-associated mutations and *BCR-ABL1* rearrangements.

We retrospectively reviewed the medical records of 38 CMML patients diagnosed according to the 2008 WHO criteria at seven university hospitals in Korea from January 2012 to July 2016. The laboratory data included a complete blood cell count with WBC differentials, BM morphology, a cytogenetic study, and a molecular genetic study including *BCR-ABL1* and the mutational status of *JAK2* V617F, *JAK2* exon12, *CALR*, and *MPL*. In addition, an analysis for MPN-associated mutations was performed for 11 patients with available archival BM specimens. Allele-specific real-time PCR was used to detect *JAK2* V617F. Sequencing was performed for both *JAK2* exon12 and *MPL* W515. PCR/fragment analysis and sequencing was conducted for *CALR* exon 9. This study was approved by the Institutional Review Board of Ajou University Hospital.

Of the 38 CMML patients, three had <10% PB monocytes and eight had MPN-associated mutations according to the 2016 WHO criteria, which precluded their classification as CMML. These 11 patients were designated as the 2016 WHO others group and the remaining 27 as the 2016 WHO CMML group. The Mann-Whitney U test was used to compare the groups. Data were analyzed using SPSS 12.0.1 for Windows (SPSS Inc., Chicago, IL, USA). $P<0.05$ was considered statistically significant.

Table 1 shows the patients' demographic and laboratory characteristics. The significant difference between the two groups in platelet count and monocyte (%) level indicates that the 2016 WHO criteria offer a more homogenous and defined classification of CMML than the 2008 WHO criteria.

The 2016 WHO CMML group was subdivided according to the 2016 WHO classification as follows: 17 (63%) as CMML-0 (<2% blasts in PB and <5% blasts in BM), five (18.5%) as CMML-1 (2–4% blasts in PB and/or 5–9% blasts in BM), and five (18.5%) as CMML-2 (5–19% blasts in PB, 10–19% blasts in BM, and/or presence of any Auer rods). According to the 2008 WHO classification, the total samples were subdivided into 32 CMML-1 and six CMML-2 patients. Sixteen patients (59%) in the 2016 WHO CMML group had proliferative type CMML (WBC counts $\geq 13\times10^9$/L), and 11 (41%) had dysplastic type CMML (WBC counts $<13\times10^9$/L). Most dysplastic type patients (9/11) had CMML-0. The dysplastic type within CMML-0 is known to have a better prognosis than other CMML subgroups [6]. Mild to moderate reticulin fibrosis in BM was present in 11 (69%) of the 16. None of the patients had a previous history of MPN.

Of the 38 CMML patients, MPN-associated mutations were found in eight (31%) of the 26 tested for the presence of ≥1 mutation (Table 2). All 27 patients in the 2016 WHO CMML group tested negative for *BCR-ABL1*. Utilization of MPN-associated mutations at diagnosis was very low, except for *JAK2* V617F (Table 2). Clonal cytogenetic abnormalities, including trisomy 8, -Y, -7/del(7q), and trisomy 21, were present in eight (30%) of the 2016 WHO CMML group and three (27%) of the 2016 WHO others group. No patient had a chromosomal rearrangement involving 8p11, corresponding to the *FGFR1* rearrangement, or t(8;9)(p22;p24.1), corresponding to *PCM-JAK2*. Clonal cytoge-

Table 1. Clinical and laboratory characteristics of patients diagnosed as having chronic myelomonocytic leukemia according to the WHO criteria

	2008 WHO (N=38)	2016 WHO CMML (N=27)	2016 WHO others (N=11)	*P**
Sex (M:F)	2.8:1	2.9:1	2.7:1	
Age	71±12†	70±12†	73±13†	0.326
WBC (×10⁹/L)	22.1 (36.6)‡	19.7 (58.2)‡	35.0 (18.8)‡	0.267
Hb (g/L)	96±21†	94±19†	101±25†	0.411
PLT (×10⁹/L)	147 (201)‡	114 (133)‡	263 (139)‡	0.005
Blast (%)	1.2±2.1†	0.9±1.9†	1.8±2.8†	0.075
MON (%)	22±10†	24±9†	14±7†	0.003
MON (×10⁹/L)	4.1 (7.9)‡	3.8 (10.8)‡	4.3 (3.4)‡	0.664
EOS (%)	1 (2)‡	1 (1)‡	1 (3)‡	0.671
BASO (%)	0.9±1.8†	0.7±1.9†	1.4±1.8†	0.109
NRBC§	0.6±1.1†	0.6±1.2†	0.6±0.7†	0.302
M:E ratio	6.9±5.6†	6.7±6.2†	7.4±4.2†	0.215
BM blast (%)	3.0 (3.0)‡	2.8 (7.0)‡	2.2 (2.2)‡	0.595
MON (%)	7.9±6.2†	8.9±6.7†	5.4±4.0†	0.122

*The Mann-Whitney U test was used to compare the 2016 WHO CMML group and the 2016 WHO others group; †Values are presented as mean±SD; ‡Values are presented as median (interquartile range); §observed per 100 WBCs.
Abbreviations: CMML, chronic myelomonocytic leukemia; M:F, male to female ratio; WBC, white blood cells; PLT, platelets; MON, monocytes; EOS, eosinophils; BASO, basophils; NRBC, nucleated red blood cells; M:E, myeloid to erythroid; BM, bone marrow.

Table 2. Myeloproliferative neoplasm-associated mutations in 38 patients diagnosed as having chronic myelomonocytic leukemia according to the 2008 WHO classification

Mutations	Test ordered at diagnosis (%)	Positive/Tested (%)
JAK2 V617F	19/38 (50)	7/26 (27)
JAK2 exon 12	1/38 (3)	0/12 (0)
CALR	4/38 (11)	0/15 (0)
MPL	4/38 (11)	1/14 (7)

https://doi.org/10.3343/alm.2018.38.5.481

netic changes have been demonstrated in 20–40% of patients with CMML, but none are specific [3, 7].

Recent studies have shown that at least one pathogenic mutation can be identified in >90% of CMML patients [8, 9]. Gene mutations frequently observed in CMML include *TET2, SRSF2, ASXL1, RAS, CBL1*, and *RUNX1*. Next-generation sequencing for pathogenic mutations may be helpful for establishing a correct diagnosis in diagnostically difficult cases of CMML. In addition, a robust multiparameter flow cytometry assay could distinguish CMML from a diagnosis of reactive monocytosis and myeloid malignancies in patients with a borderline or increased monocyte count by detecting an increase in the fraction of classical CD14+/CD16- cells among the circulating monocytes [10].

Our data show that the rate of tests ordered for the detection of MPN-associated mutations at initial diagnosis is very low, except for the testing of *JAK2* V617F. This was mainly because the national health insurance used to only cover *JAK2* V617F testing. Currently, the national health insurance system in Korea covers all four MPN-associated mutations; thus, the usage rate is expected to increase. In addition, hematologists and hematopathologists need to make an effort to adhere to the 2016 WHO classification.

In conclusion, the 2008 WHO criteria may result in over-diagnosis of CMML compared with the 2016 WHO criteria. More aggressive use of molecular tests, including MPN-associated mutation analyses, and a more sophisticated clinical or morphological evaluation are necessary in order to achieve an accurate CMML diagnosis.

Authors' Disclosures of Potential Conflicts of Interest

No potential conflicts of interest relevant to this article were reported.

Acknowledgment

This study was supported by a research grant from Inha University.

REFERENCES

1. Itzykson R, Duchmann M, Lucas N, Solary E. CMML: clinical and molecular aspects. Int J Hematol 2017;105:711-9.
2. Santini V, Allione B, Zini G, Gioia D, Lunghi M, Poloni A, et al. A phase II, multicenter trial of decitabine in higher-risk chronic myelomonocytic leukemia. Leukemia 2018;32:413-8.
3. Arber DA, Orazi A, Hasserjian R, Thiele J, Borowitz MJ, Le Beau MM, et al. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. Blood 2016;127:2391-405.
4. Orazi A, Bennett JM, et al. Chronic myelomonocytic leukemia. In: Swerdlow SH, Campo E, et al. eds. WHO classification of tumours of haematopoietic and lymphoid tissues. Revised 4th ed. Lyon: International Agency for Research on Cancer, 2017:82-6.
5. Orazi A, Bennett JM, et al. Chronic myelomonocytic leukemia. In: Swerdlow SH, Campo E, et al. eds. WHO classification of tumours of haematopoietic and lymphoid tissues. 4th ed. Lyon: International Agency for Research on Cancer, 2008:76-9.
6. Schuler E, Schroeder M, Neukirchen J, Strupp C, Xicoy B, Kündgen A, et al. Refined medullary blast and white blood cell count based classification of chronic myelomonocytic leukemias. Leuk Res 2014;38:1413-9.
7. Patnaik MM and Tefferi A. Cytogenetic and molecular abnormalities in chronic myelomonocytic leukemia. Blood Cancer J 2016;6:e393.
8. Elena C, Gallì A, Such E, Meggendorfer M, Germing U, Rizzo E, et al. Integrating clinical features and genetic lesions in the risk assessment of patients with chronic myelomonocytic leukemia. Blood 2016;128:1408-17.
9. Reinig E, Yang F, Traer E, Arora R, Brown S, Rattray R, et al. Targeted next-generation sequencing in myelodysplastic syndrome and chronic myelomonocytic leukemia aids diagnosis in challenging cases and identifies frequent spliceosome mutations in transformed acute myeloid leukemia. Am J Clin Pathol 2016;145:497-506.
10. Selimoglu-Buet D, Wagner-Ballon O, Saada V, Bardet V, Itzykson R, Bencheikh L, et al. Characteristic repartition of monocyte subsets as a diagnostic signature of chronic myelomonocytic leukemia. Blood 2015;125:3618-26.