Immunopathologia Persa
http immunopathol.com
DOI:10.34172/ipp.2025.43799

CD34+CD38-stem cells and CD34+CD38+progenitor cells as markers of chemotherapy response in acute myeloid leukemia patients

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Received 25 Nov. 2024
Revised: 12 Feb. 2025
Accepted 8 Mar. 2025
ePublished 22 May 2025

Keywords: Acute myeloid leukemia, Complete remission, Flow cytometry, CD34, CD38, Induction chemotherapy, Incomplete remission, Leukemic stem cells

Abstract

Introduction: CD34+CD38-stem cells were identified as the most related markers to acute myeloid leukemia (AML) progression, resistance, and relapse. However, there is still a lack of published data identifying the level of CD34 and CD38 during induction chemotherapy and after complete remission.
Objectives: This study aimed to evaluate the levels of CD34+CD38+progenitor cells and CD34+CD38-stem cells in AML patients at diagnosis and after induction chemotherapy.
Patients and Methods: This is a prospective cohort study. Both CD34 and CD38 cell markers were identified using flow cytometry in newly diagnosed AML patients and after induction chemotherapy.
Results: Forty newly diagnosed AML patients (27 males and 13 females) were followed up after induction chemotherapy. Results revealed a statistically significant decline ($P \leq 0.05$) in CD34+CD38-stem cell levels as well as in CD34+CD38+progenitor cell levels in AML patients who achieved complete or incomplete remission compared to newly diagnosed AML patients. Besides, age and CD34+CD38-stem cells exhibited a statistically significant positive correlation at diagnosis.
Conclusion: Our study showed CD34+CD38-stem cells are associated with disease progression and poor survival in AML patients. We concluded that, CD34 and CD38 are promising follow-up markers for AML patients on induction chemotherapy. In order to keep AML patients in full remission and increase their chances of survival, medications that target leukemic stem cells (LSCs) should be used in conjunction with the usual chemotherapy that targets blasts.

Citation: Fergany A, Hassanein KM, Abdel Hameed MR, Kamel AM, Zahran AM. CD34+CD38-stem cells and CD34 + CD38 + progenitor cells as markers of chemotherapy response in acute myeloid leukemia patients . Immunopathol Persa. 2025;11(2):e43799. DOI:10.34172/ipp.2025.43799.

Introduction

The hallmark of acute myeloid leukemia (AML) is the clonal development of myeloid leukemia blasts in the bone marrow and peripheral blood (1-3). About 90% of adult cases of acute leukemia are AML, and their prevalence rises with age (4). Patients with AML often have a bad prognosis (the 5-year overall survival for those who are over 60 years old is 20%) (5). Persistent neutropenia from combination chemotherapy for AML treatment raises the risk of opportunistic infections, mostly by resistant candida species; hence there is a high demand for novel therapeutic approaches (6,7).

The chromosomal rearrangements and numerous gene mutations that result in AML are caused by the transformation of hematopoietic stem or progenitor cells (8,9). Leukemic stem cells (LSCs) are considered as a subset of cells in AML that are essential to disease onset and progression (10). An undifferentiated state, self-renewal, and resistance to drugs are among the stemness characteristics that define LSCs. Experts believe that LSCs contribute to the poor prognosis and illness recurrence in AML patients (11-15).

CD34+CD38+ and CD34+CD38- subpopulation are arguments (16-20), although many studies demonstrated the association of CD34 expression with AML patient outcomes, controversy still exists (20, 21). Previous studies have shown the association of CD34+CD38- with poor prognosis; however, data identifying the level of CD34 and CD38 after induction chemotherapy are still limited.

Key point

- CD34+CD38- stem cells are associated with AML patients' disease progression and poor survival.
- Medications that target leukemic stem cells should be used in conjunction with the usual chemotherapy that targets blasts.

Objectives

This study aimed to evaluate and compare the level of CD34+CD38-stem cells and CD34+CD38+progenitor cells in patients with AML at diagnosis and after induction chemotherapy.

Patients and Methods

Study design and setting

This is a prospective cohort study. This study was conducted in the clinical pathology department at South Egypt cancer institute (SECI), in cooperation with clinical hematology unit, faculty of medicine, Assiut university, Egypt. This research included all newly diagnosed adult AML patients hospitalized between December 2022 and September 2023. Patients were followed up after chemotherapy until complete remission or death. Inclusion criteria were; the newly diagnosed AML patients (AML-ND) before intensive induction chemotherapy aged from 18 years old. Exclusion criteria: AML patients on low-intensity chemotherapy courses, recurrent AML patients, AML M3 (acute promyelocytic leukemia), and AML patients with prior hematologic malignancies such as myelodysplastic syndrome, myeloproliferative neoplasms. The patients treated with the following chemotherapy regimen; cytarabine (100 mg/m^2/day for 5–7 days) and idarubicin (12 mg/m^2/day) (6).

Patients' diagnosis

The diagnosis was conducted using the bone marrow examination, complete blood count, clinical presentation, immunophenotyping and cytogenetic studies. Immunophenotyping using monoclonal antibodies for staining the blast cells using flowcytometry FACSCanto II (BD Bioscience, USA). In order to confirm or refute the diagnosis of AML, markers were employed including; CD2, CD3, CD7, CD13, CD14, CD33, CD36, MPO, CD45, CD117, CD 41, CD61, and CD64, CD4, CD5, CD10, CD19, CD34 and HLA-DR. All monoclonal antibodies which were brought from Beckman Coulter, USA. The FACS Canto II and Navios EX 3L/10C (S.N.BF22S40) (Beckman Coulter, USA) flow cytometry and the FACS Diva and Kaluza analysis software were used respectively. The 2010 European Leukemia Network (ELN) guideline was followed when defining the response to chemotherapy (22). This study includes 40 newly diagnosed (AML-ND) patients, 13 incomplete remission (AML-IR) patients, and 12 complete remission (AML-CR) patients.

For detection of LSC population, 2 mL of fresh blood were collected from patients with AML to quantify the LSCs with a Becton Dickinson *FACSCalibur*™ flow cytometer (San Jose, CA, USA). Following the staining of 100 µL of blood with 10 µL of phycoerythrin (PE) conjugated anti-CD38 and 10 µL of fluorescein isothiocyanate conjugated anti-CD34 (Bioscience, USA), the samples were allowed to sit at room temperature for 15 minutes. Then red blood cells (RBCs) were lysed using RBCs lysis buffer, then washed using phosphate buffered saline following the incubation period. Accordingly, Kaluza analysis software, and FACS acquisition and analysis were conducted to analyze the expression level of CD34 and CD38 to detect LSC population. Meanwhile, the CD34+CD38+progenitor cells and CD34+CD38-stem cells represented the proportion of all blast cells (Figure 1).

Statistical analysis

Median ± interquartile range (IQR), was conducted to express quantitative data. The quantitative data's normality was confirmed using the Shapiro-Wilk test. The Kruskal-Wallis test, pairwise comparison test, and Mann-Whitney U test were employed to assess the group differences. Spearman's correlation coefficient test was conducted to establish correlation between non-normally distributed data. All statistical tests employed in the investigation were conducted at a significance level of 5%. Version 26 of IBM SPSS Statistics for Windows was conducted to analyze the data.

Results

Table 1 shows the laboratory and demographic data of

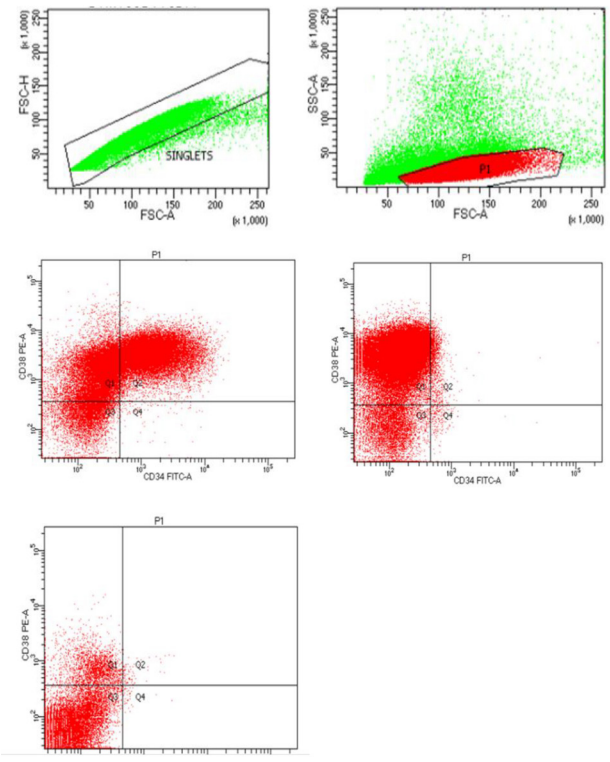

Figure 1. Flowcytometry analysis of LSC and progenitor cells in three representative cases.

Table 1. Laboratory and demographic data of patient's groups

Parameter	AML-ND (n=40)	AML-IR (n=13)	AML-CR (n=12)
Age (year)	35.5 (21)	30 (17.5)	29.5 (16.75)
Blasts %	68.8 (13.93)	8 (14)	2.7 (0.82)
WBCs (10^3/μL)	44.75 (67.63)	6.1 (5.19)	6.35 (1.5)
Platelets (10^3/μL)	44.5 (64.33)	294.36 (237)	414 (149.5)
Hemoglobin (g/dL)	8.7 (2)	10.4 (2.35)	10.78 (1.42)
Demographic data of AML-ND (n=40)			
Fever	40%		
Hepatomegaly	27.5%		
Hepatosplenomegaly	12.5%		
Anemia	40%		
Other	47.5%		

All data is expressed as median (IQR). Demographic data is expressed in percentage.
Abbreviations: Newly diagnosed AML (AML-ND), complete remission (AML-CR), and incomplete remission (AML-IR).

the patient groups. We followed up 40 AML-ND patients (27 males and 13 females) after induction chemotherapy. Thirteen patients (32.5%) were in incomplete remission (AML-IR). Twelve patients (30%) achieved complete remission (AML-CR). The rest of the patients (37.5%) died before completing the induction chemotherapy.

Comparison of CD34+CD38-stem cells and CD34 + CD38 + progenitor cells levels among AML-ND, AML-IR and AML-CR patient groups

Figure 2 shows a comparison between the level of CD34+CD38-stem cells within the three groups (AML-ND, AML-IR and AML-CR). Results revealed a statistically significant decline ($P < 0.001$) in CD34+CD38-stem cell levels expression after induction chemotherapy compared to their levels at diagnosis. However, there was no statistically significant change in CD34+CD38-stem cell levels of expression between AML-IR and AML-CR groups.

Figure 3 shows a comparison between levels of CD34+CD38+progenitor cells within the three groups (AML-ND, AML-IR and AML-CR). Results showed a statistically significant decline ($P = 0.007$) in CD34+/CD38+progenitor cells after induction chemotherapy compared to their levels at diagnosis. However, there was no statistically significant change in CD34+CD38+progenitor cells level of expression between AML-IR and AML-CR groups.

Correlation between CD34+CD38-stem cells, CD34 + CD38+progenitor cells levels and laboratory data among patient groups AML-ND, AML-IR, and AML-CR

In the AML-ND group, there was a statistically significant positive correlation between age and CD34+CD38-stem cells, and a statistically significant negative correlation between CD34+CD38+progenitor cells and hemoglobin

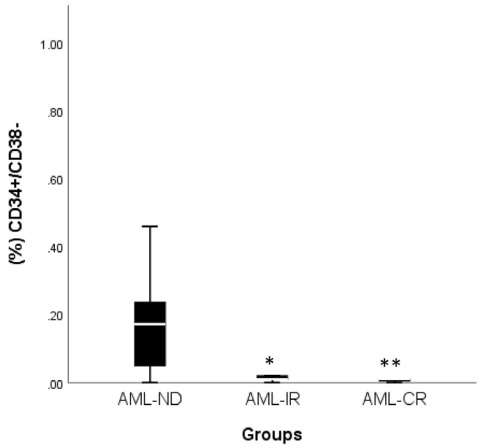

Figure 2. A comparison among levels of CD34+CD38-stem cell expression within the AML-ND, AML-IR, and AML-CR patient groups. * indicates a statistically significant difference (P≤ 0.05) between AML-IR group and AML-ND group. ** indicates a statistically significant difference ($P \leq 0.05$) between AML-CR group and AML-ND group. Abbreviations: Newly diagnosed AML (AML-ND), complete remission (AML-CR), and incomplete remission (AML-IR).

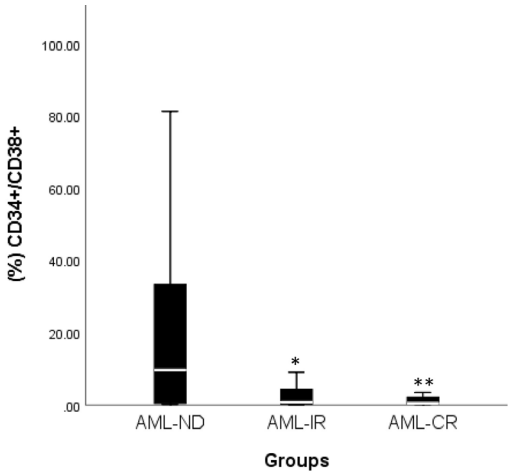

Figure 3. A comparison among levels of CD34+CD38+progenitor cell expression within AML-ND, AML-IR, and AML-CR patient groups. The Kruskal-Wallis test, pairwise comparison test was used. * indicates a statistically significant difference (P≤ 0.05) between AML-IR group and AML-ND group. ** indicates a statistically significant difference ($P \leq 0.05$) between AML-CR group and AML-ND group. Abbreviations: Newly diagnosed AML (AML-ND), complete remission (AML-CR), and incomplete remission (AML-IR).

levels. In the AML-IR group, CD34+CD38+progenitor cells exhibited a statistically significant positive correlation with monocytes and statistically significant negative correlation with myelocytes. Moreover, promyelocytes and CD34+CD38-stem cells had a statistically significant negative correlation, while hemoglobin and CD34+CD38-stem cells had a statistically significant positive correlation. In the AML-CR group, CD34+CD38-stem cells and hemoglobin had a statistically significant negative correlation as shown in Table 2.

Discussion

Leukemic stem cells are believed to be strong predictors

Table 2. Correlations between CD34+CD38+progenitor cells and CD34+CD38-stem cells, and laboratory parameters

Parameter	AML-ND (N=40)		AML-IR (N=13)		AML-CR (N=12)	
	CD34+ CD38-	CD34+ CD38+	CD34+ CD38-	CD34+ CD38+	CD34+ CD38-	CD34+ CD38+
Age (year)	r = 0.359* *P* = 0.023	r = 0.146 *P* = 0.312	r = 0.216 *P* = 0.479	r = 0.135 *P* = 0.661	r = -0.5 *P* = 0.098	r = 0.164 *P* = 0.651
Blasts %	r = -0.058 *P* = 0.723	r = -0.013 *P* = 0.936	r = 0.026 *P* = 0.932	r = 0.141 *P* = 0.646	r = -0.077 *P* = 0.813	r = 0.057 *P* = 0.862
WBCs (10^3/μL)	r = -0.067 *P* = 0.679	r = -0.112 *P* = 0.491	r = 0.414 *P* = 0.16	r = 0.08 *P* = 0.795	r = 0.105 *P* = 0.746	r = -0.120 *P* = 0.711
Platelets (10^3/μL)	r = -0.092 *P* = 0.574	r = -0.184 *P* = 0.255	r = -0.185 *P* = 0.545	r = -0.477 *P* = 0.1	r = -0.288 *P* = 0.365	r = -0.222 *P* = 0.489
Hemoglobin (g/dL)	r = -0.068 *P* = 0.677	r = -0.037 *P* = 0.019*	r = 0.555* *P* = 0.049	r = 0.121 *P* = 0.693	r = -0.68* *P* = 0.015	r = -0.519 *P* = 0.084
Promyelocytes%	r = -0.241 *P* = 0.134	r = -0.193 *P* = 0.232	r = -0.679* *P* = 0.011	r = -0.465 *P* = 0.109	r = -0.157 *P* = 0.625	r = 0.049 *P* = 0.879
Myelocytes %	r = 0.043 *P* = 0.792	r = -0.01 *P* = 0.95	r = -0.024 *P* = 0.938	r = -0.561* *P* = 0.046	r = 0.149 *P* = 0.645	r = -0.053 *P* = 0.87
Neutrophils %	r = 0.144 *P* = 0.377	r = -0.026 *P* = 0.875	r = 0.332 *P*= 0.268	r = 0.003 *P* = 0.993	r = -0.545 *P* = 0.067	r = -0.069 *P* = 0.831
Lymphocytes%	r = 0.152 *P* = 0.349	r = 0.006 *P* = 0.972	r = -0.053 *P* = 0.864	r = 0.141 *P* = 0.647	r = -0.144 *P* = 0.656	r = 0.2 *P* = 0.534
Monocyte%	r = 0.003 *P* = 0.985	r = 0.128 *P* = 0.432	r = 0.306 *P* = 0.309	r = 0.796** *P* = 0.001	r = 0.129 *P* = 0.69	r = -0.029 *P* = 0.928
Eosinophils%	r = -0.133 *P* = 0.413	r = -0.111 *P* = 0.496	r = -0.427 *P* = 0.145	r = -0.097 *P* = 0.751	r = -0.173 *P* = 0.591	r = 0.401 *P* = 0.196
Basophils%	r = -0.063 *P* = 0.701	r = -0.211 *P* = 0.19	r = 0.201 *P* = 0.511	r = 0.127 *P* = 0.679	-	-

Data is expressed as Spearman's correlation coefficient (r). * represents a statistical significance at the level of 0.05. ** represents statistical significance at the level of 0.01. Abbreviations: Newly diagnosed AML (AML-ND), complete remission (AML-CR), and incomplete remission (AML-IR).

of AML clinical outcomes and is associated with poorer outcomes, such as remission and survival, in AML patients (19,23). CD34+CD38-stem cells may be the reason for AML treatment failure because of their chemotherapy-resistant characteristics (21,23). Besides, it is critical to determine whether putative LSCs will withstand therapy for the purpose of therapeutic treatment and patient survival (24,25). More knowledge of LSCs will enable the development of more potent treatments (14). This study aimed to evaluate the levels of CD34+CD38-stem cells in AML patients at diagnosis and after induction chemotherapy.

In this study, 65 AML patients' samples were included. In terms of demographic information, patients enrolled in the study varied in age from 18 to 61 years old, with a mean age of 35.5 years. Pervious research carried out in Upper Egypt on 170 patients with AML revealed that the patients were between the ages of 18 and 69, with a mean age of 49. The ratio of male to females was a modest 1:1.36 (6).

Figures 2 and 3 compare the levels of CD34 +CD38+progenitor cells, and CD34+CD38-stem cells within AML-ND, AML-IR and AML-CR patient groups. Results revealed a statistically significant decline in CD34+CD38+progenitor cell, and CD34+CD38-stem cell levels in AML-IR and AML-CR groups compared to AML-ND group. This result indicates the efficacy of induction chemotherapy to eradicate CD34+CD38+progenitor cells, as well as CD34+CD38-stem cells. Hence, these flow cytometry markers are considered to be a helpful, independent and sensitive follow up and predictor markers for chemotherapy outcome (20,27,28). However, no statistically significant difference was observed in levels of CD34+CD38+progenitor cell or CD34+CD38-stem cell among AML-IR and AML-CR groups. This result supports the growing-up hypothesis that suggests a small fraction within LSCs (but not the bulk of them) is responsible for resistance and relapse and requires additional chemotherapy cycles (16,20,27,28).

Table 2 shows a statistically significant positive correlation between age and CD34+CD38-stem cells, which indicates a poor response with older patients in the AML-ND group. A pervious study results showed that age is a reliable predictive factor on its own in AML and CD38 is decreased in elderly patients (older than 60 years) (20). In addition, CD34+CD38+progenitor cells and hemoglobin showed a statistically significant negative correlation. In AML-IR group, CD34+CD38+progenitor cells exhibited a statistically significant positive correlation with monocytes and statistically significant negative correlation with myelocytes. This result indicates the improvement of maturation in bone marrow microenvironment of AML after induction of chemotherapy despite not achieving complete remission (16). In addition, CD34+CD38-stem cells and hemoglobin level showed a statistically significant correlation in both AML-IR and AML-CR groups.

Low-hemoglobin levels are associated with a number of detrimental health effects, such as decreased muscle strength and difficulty walking, as well as an increase in weariness, sadness and a loss in quality of life (29).

Conclusion

This study proved that flow cytometry markers CD34 and CD38 are promising helpful, follow up, and predictor markers for AML patients on induction chemotherapy. CD34+CD38-stem cells in AML patients are associated with disease progression and poor survival. In order to keep AML patients in full remission and increase their chances of survival, medications that target LSCs should be used in conjunction with the usual chemotherapy that targets blasts.

Limitations of the study

This study has some limitations because of the small sample sizes in the follow up AML-IR and AML-CR groups. Extended follow-up research is required to prove the presence and contribution of CD34+CD38- percentage in AML patients with chemotherapy resistance and AML relapse.

Acknowledgements

The authors gratefully acknowledge the Regional Ethical Committee of the Faculty of Medicine at Assiut University for authorizing this research project.

Authors' contribution

Conceptualization: All authors.
Data curation: Aya Fergany, Muhamad R Abdel Hameed, and Asmaa M. Zahran.
Formal analysis: Aya Fergany, and Asmaa M. Zahran.
Investigation: All authors.
Methodology: Aya Fergany, Muhamad R Abdel Hameed, and Asmaa M. Zahran.
Project administration: Khaled M Hassanein, Muhamad R Abdel Hameed, Ayat M. Kamel, and Asmaa M. Zahran.
Resources: Aya Fergany, Muhamad R Abdel Hameed, and Asmaa M. Zahran.
Software: Asmaa M. Zahran.
Supervision: Khaled M Hassanein, Muhamad R Abdel Hameed, Ayat M. Kamel, and Asmaa M. Zahran.
Validation: Aya Fergany, and Asmaa M. Zahran.
Visualization: Aya Fergany, and Asmaa M. Zahran.
Writing–original draft: Aya Fergany.
Writing–review & editing: All authors.

Conflicts of interest

The authors declare that they have no competing interests.

Ethical issues

This study adhered to the principles outlined in the Declaration of Helsinki and was approved by the Ethics Committee of the Faculty of Medicine at Assiut University (ethical code 17200798) for Aya Fergany's doctoral thesis. Prior to any intervention, all participants provided written informed consent. Ethical issues (including plagiarism, data fabrication, double publication) have been completely observed by the authors.

Funding/Support

None.

References

1. Estey E, Döhner H. Acute myeloid leukaemia. Lancet. 2006;368:1894-907. doi: 10.1016/s0140-6736(06)69780-8.
2. Bashi MA, Ad'hiah AH. Interleukin-37 gene expression is down-regulated in patients with acute myeloid leukemia and shown to be affected by CD14 and HLA-DR immunophenotypes. Cytokine. 2023;171:156368. doi: 10.1016/j.cyto.2023.156368.
3. Nix NM, Price A. Optimizing use of newly approved agents for acute myeloid leukemia. J Adv Pract Oncol. 2019;10:9-18. doi: 10.6004/jadpro.2019.10.8.13.
4. Tao Q, Pan Y, Wang Y, Wang H, Xiong S, Li Q, et al. Regulatory T cells-derived IL-35 promotes the growth of adult acute myeloid leukemia blasts. Int J Cancer. 2015;137:2384-93. doi: 10.1002/ijc.29563.
5. Eriksson M, Peña-Martínez P, Ramakrishnan R, Chapellier M, Högberg C, Glowacki G, et al. Agonistic targeting of TLR1/TLR2 induces p38 MAPK-dependent apoptosis and NFκB-dependent differentiation of AML cells. Blood Adv. 2017;1:2046-57. doi: 10.1182/bloodadvances.2017006148.
6. Sayed SA, Hassan EAB, Abdel Hameed MR, Agban MN, Mohammed Saleh MF, Mohammed HH, et al. Ketorolac-fluconazole: a new combination reverting resistance in *Candida albicans* from acute myeloid leukemia patients on induction chemotherapy: in vitro study. J Blood Med. 2021;12:465-74. doi: 10.2147/jbm.S302158.
7. Elgendy SG, Abdel Hameed MR, El-Mokhtar MA. Tigecycline resistance among Klebsiella pneumoniae isolated from febrile neutropenic patients. J Med Microbiol. 2018;67:972-5. doi: 10.1099/jmm.0.000770.
8. Estey EH. Acute myeloid leukemia: 2012 update on diagnosis, risk stratification, and management. Am J Hematol. 2012;87:89-99. doi: 10.1002/ajh.22246.
9. Chávez-González A, Dorantes-Acosta E, Moreno-Lorenzana D, Alvarado-Moreno A, Arriaga-Pizano L, Mayani H. Expression of CD90, CD96, CD117, and CD123 on different hematopoietic cell populations from pediatric patients with acute myeloid leukemia. Arch Med Res. 2014;45:343-50. doi: 10.1016/j.arcmed.2014.04.001.
10. Hope KJ, Jin L, Dick JE. Acute myeloid leukemia originates from a hierarchy of leukemic stem cell classes that differ in self-renewal capacity. Nat Immunol. 2004;5:738-43. doi: 10.1038/ni1080.
11. Zhou J, Chng WJ. Identification and targeting leukemia stem cells: the path to the cure for acute myeloid leukemia. World J Stem Cells. 2014;6:473-84. doi: 10.4252/wjsc.v6.i4.473.
12. Han H, Byun JM, Shin DY, Yoon SS, Koh Y, Hong J, et al. Leukemic stem cell phenotype is associated with mutational profile in acute myeloid leukemia. Korean J Intern Med. 2021;36:401-12. doi: 10.3904/kjim.2020.014.
13. Thomas D, Majeti R. Biology and relevance of human acute myeloid leukemia stem cells. Blood. 2017;129:1577-85. doi: 10.1182/blood-2016-10-696054.
14. Pollyea DA, Gutman JA, Gore L, Smith CA, Jordan CT. Targeting acute myeloid leukemia stem cells: a review and principles for the development of clinical trials. Haematologica. 2014;99:1277-84. doi: 10.3324/haematol.2013.085209.
15. Pollyea DA, Jordan CT. Therapeutic targeting of acute myeloid leukemia stem cells. Blood. 2017;129:1627-35. doi: 10.1182/blood-2016-10-696039.
16. Zeijlemaker W, Grob T, Meijer R, Hanekamp D, Kelder A, Carbaat-Ham JC, et al. CD34+CD38- leukemic stem cell frequency to predict outcome in acute myeloid leukemia. Leukemia. 2019;33:1102-12. doi: 10.1038/s41375-018-0326-3.
17. Costello RT, Mallet F, Gaugler B, Sainty D, Arnoulet C, Gastaut

JA, et al. Human acute myeloid leukemia CD34+/CD38- progenitor cells have decreased sensitivity to chemotherapy and Fas-induced apoptosis, reduced immunogenicity, and impaired dendritic cell transformation capacities. Cancer Res. 2000;60:4403-11.
18. Ishikawa F, Yoshida S, Saito Y, Hijikata A, Kitamura H, Tanaka S, et al. Chemotherapy-resistant human AML stem cells home to and engraft within the bone-marrow endosteal region. Nat Biotechnol. 2007;25:1315-21. doi: 10.1038/nbt1350.
19. Terwijn M, Zeijlemaker W, Kelder A, Rutten AP, Snel AN, Scholten WJ, et al. Leukemic stem cell frequency: a strong biomarker for clinical outcome in acute myeloid leukemia. PLoS One. 2014;9:e107587. doi: 10.1371/journal.pone.0107587.
20. Plesa A, Dumontet C, Mattei E, Tagoug I, Hayette S, Sujobert P, et al. High frequency of CD34+CD38-/low immature leukemia cells is correlated with unfavorable prognosis in acute myeloid leukemia. World J Stem Cells. 2017;9:227-34. doi: 10.4252/wjsc.v9.i12.227.
21. van Rhenen A, Feller N, Kelder A, Westra AH, Rombouts E, Zweegman S, et al. High stem cell frequency in acute myeloid leukemia at diagnosis predicts high minimal residual disease and poor survival. Clin Cancer Res. 2005;11:6520-7. doi: 10.1158/1078-0432.Ccr-05-0468.
22. Döhner H, Estey EH, Amadori S, Appelbaum FR, Büchner T, Burnett AK, et al. Diagnosis and management of acute myeloid leukemia in adults: recommendations from an international expert panel, on behalf of the European LeukemiaNet. Blood. 2010;115:453-74. doi: 10.1182/blood-2009-07-235358.
23. Eppert K, Takenaka K, Lechman ER, Waldron L, Nilsson B, van Galen P, et al. Stem cell gene expression programs influence clinical outcome in human leukemia. Nat Med. 2011;17:1086-93. doi: 10.1038/nm.2415.
24. Costello R, Mallet F, Chambost H, Sainty D, Arnoulet C, Gastaut JA, et al. The immunophenotype of minimally differentiated acute myeloid leukemia (AML-M0): reduced immunogenicity and high frequency of CD34+/CD38- leukemic progenitors. Leukemia. 1999;13:1513-8. doi: 10.1038/sj.leu.2401519.
25. Zahran AM, Aly SS, Rayan A, El-Badawy O, Fattah MA, Ali AM, et al. Survival outcomes of CD34+CD38-LSCs and their expression of CD123 in adult AML patients. Oncotarget. 2018;9:34056-65. doi: 10.18632/oncotarget.26118.
26. Chen X, Othus M, Wood BL, Walter RB, Percival MM, Becker PS, et al. Flow cytometric demonstration of decrease in bone marrow leukemic blasts after 'Day 14' without further therapy in acute myeloid leukemia. Leuk Lymphoma. 2017;58:2717-9. doi: 10.1080/10428194.2017.1307359.
27. Bonnet D, Dick JE. Human acute myeloid leukemia is organized as a hierarchy that originates from a primitive hematopoietic cell. Nat Med. 1997;3:730-7. doi: 10.1038/nm0797-730.
28. Becker MW, Jordan CT. Leukemia stem cells in 2010: current understanding and future directions. Blood Rev. 2011;25(2):75-81. doi: 10.1016/j.blre.2010.11.001.
29. Fukushima T, Nakano J, Ishii S, Natsuzako A, Kawachi H, Sakamoto J, et al. Influence of hemoglobin level on muscle and physical functions, activities of daily living, and quality of life in patients with hematological malignancies. Integr Cancer Ther. 2019;18:1534735419842196. doi: 10.1177/1534735419842196.