Two Cases of Acute Erythroid Leukemia Presenting with Marked Macrocytic Anemia, Reticulocytosis and Hemolysis

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Abstract

Case 1. The laboratory findings of a hematological analysis of a 53-year-old woman with palpitations and dyspnea revealed the following: red blood cell (RBC) count: $9.4 \times 10^5/\mu\text{L}$ with 60.0% reticulocytes; Hb: 3.7 g/dL; mean corpuscular volume (MCV): 124.5 fL; white blood cell (WBC) count: $2,800/\mu\text{L}$ with 10.0% myeloblasts. Case 2. Similarly, a 42-year-old man with dizziness had a RBC count of $1.63 \times 10^6/\mu\text{L}$ with 24.0% reticulocytes, an Hb level of 6.0 g/dL, an MCV of 120.2 fL and a WBC count of $3,100/\mu\text{L}$ with 4.0% myeloblasts. Bone marrow aspirates in both patients confirmed a diagnosis of acute erythroid leukemia (AEL), which can present as marked macrocytic anemia with an MCV in excess of 120 fL and hemolysis.

Key words: acute erythroid leukemia, macrocytic anemia, hemolysis

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Introduction

Macrocytic anemia, defined as a mean corpuscular volume (MCV) >100 fL can be caused by multiple factors, including interference with DNA synthesis due to folate or cobalamin deficiency (1-4) and liver diseases, such as alcohol-induced disorders and myelodysplastic syndrome (MDS) (5). Moreover, macrocytosis is frequently drug-related, caused in particular by hydroxyurea (inhibits ribonucleotide reductase), methotrexate (inhibits dihydrofolate reductase), zidovudine (6), azathioprine or 6-mercaptopurine, capecitabine (7) or cladribine.

Peripheral reticulocytosis is another cause of macrocytic anemia. During normal erythropoiesis, erythroid precursors (orthochromatic normoblasts) extrude their nuclei while still in the bone marrow (BM), and the MCV of the resulting reticulocytes is 120-150 fL. Reticulocytes remain in the BM for approximately three days and continue to be macrocytic following their release into the peripheral blood, where they undergo remodeling with loss of their cytosol, organelles

and cell membrane. The normal MCV of peripheral reticulocytes is 103-126 fL, the mean corpuscular Hb concentration (MCHC) is 23.5-28.7 g/dL and the mean cell Hb level is 25.9-30.6 (8-11).

Hemolytic anemia (12) is caused by extravascular destruction of red blood cells, including those with intrinsic defects, such as pyruvate kinase deficiency (13), thalassemia (14) and spherocytosis, and extrinsic defects, such as liver disease and exposure to oxidant agents. Hemolytic anemia is also caused by intravascular destruction of red blood cells due to aortic stenosis or paroxysmal nocturnal hemoglobinuria.

Hemolytic anemia (12) may result from a variety of causes, typically extravascular hemolysis due to intrinsic red blood cell (RBC) defects (e.g., pyruvate kinase deficiency (13), thalassemia (14) and spherocytosis), extrinsic factors, such as liver disease or exposure to oxidizing agents, or intravascular destruction related to aortic stenosis or paroxysmal nocturnal hemoglobinuria. We herein present two unusual cases of macrocytic anemia with an MCV in excess of 120 fL with reticulocytosis and hemolysis that

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Figure. A, B. Bone marrow aspiration revealed 71.8% erythroblasts and 10.6% myeloblasts in case 1 (A, May-Grünwald-Giemsa, B, Hematoxylin and Eosin (H&E) staining, $\times 40$). Peripheral blood showing 240.0% reticulocytes and a white blood cell count with a differential count of 4.0% myeloblasts in case 2. BM aspiration revealed 68.0% erythroblasts and 5.0% myeloblasts in case 2 (D, May-Grünwald-Giemsa, E, H&E staining, $\times 40$).

were diagnosed as acute erythroid leukemia (AEL).

Case Reports

Case 1

A 53-year-old woman was referred on January, 2012 with a three-month history of palpitations and dyspnea. She was severely anemic and was therefore admitted to our hospital the following day. She had no particular medical history. A blood analysis revealed macrocytic anemia with hemolysis and pancytopenia: RBC count, $9.4 \times 10^5/\mu\text{L}$ with 60.0% reticulocytes; Hb, 3.7 g/dL; MCV, 124.5 fL; white blood cell (WBC) count, $2,800/\mu\text{L}$ with a differential count of 10.0% myeloblasts; and platelet count, $1.29 \times 10^5/\mu\text{L}$. The elevated levels of total bilirubin (2.1 mg/dL) and lactate dehydrogenase (LDH; 486 IU/L) were consistent with the findings of hemolysis. The level of haptoglobin was less than 10 mg/dL, although the type of haptoglobin was not identified. The levels of AST, ALT and γ -GTP were normal. No anti-parvovirus B19 IgM antibodies were detected. Moreover, direct and indirect Coombs tests were negative, and the vitamin B12 and folic acid levels were normal. A diagnosis of

AEL was confirmed after BM aspiration revealed 71.8% erythroblasts and 10.6% myeloblasts with a nucleated count of $4.42 \times 10^5/\mu\text{L}$ and a megakaryocyte count of $63/\mu\text{L}$ (normal range: 50-150/ μL) (Figure A, B). As the percentage of erythroblasts was 71.8%, the percentage of non-erythroid cells was 28.2%. Therefore, the proportion of 10.6% of the myeloblasts in 28.2% of the non-erythroid cells was 37.5% (Table 1). The erythroblasts exhibited megaloblastic-like abnormalities with lobulated nuclei. The neutrophils demonstrated degradation, and the number of mature neutrophils was decreased. The number of megakaryocytes was increased, and micromegakaryocytes were observed. A cytogenetic analysis revealed a normal karyotype (46, XY) in 20 cells. Thereafter, the patient was referred to another hospital for high-dose chemotherapy with BM transplantation on day 11 of admission.

Case 2

A 42-year-old man was referred to our hospital on September, 2012 with a week-long history of dizziness. He had no particular medical history. A blood analysis revealed macrocytic anemia: RBC count, $1.63 \times 10^6/\mu\text{L}$ with 240.0% reticulocytes; Hb, 6.0 g/dL; MCV, 120.2 fL; WBC count,

Table 1. Marrow Differential Cell Count in Case 1

Group	Cell type	Fraction	Reference range (%)
Cells of myelopoiesis	Myeloblasts	10.6%	0.2-2.9
	Promyelocytes	8.8%	1.5-8.4
	Myelocytes	2.6%	1.0-9.7
	Metamyelocytes	2.6%	3.6-14.6
	Band cells	0.8%	10.6-24.6
	Segmented neutrophils	1.2%	8.5-33.2
	Eosinophils	0.8%	1.2-5.3
	Basophils	0.2%	0-0.8
	Total	25.4%	50.4-70.5
Cells of erythropoiesis	Proerythroblasts	1.4%	0.2-1.3
	Basophilic erythroblasts	3.1%	0.8-6.7
	Polychromatic erythroblasts	62.1%	4.1-29.1
	Orthochromatic erythroblast	5.2%	0.1-5.7
	Total	71.8	18.4-33.8
Other cell types	Megakaryocytes	0.2%	0-0.1
	Plasma cells	0.2%	0.2-1.7
	Reticular cells	0.2%	0-3.9
	Lymphocytes	2.2%	5.0-32.6
	Monocytes	0%	0.7-6.0

Table 2. Marrow Differential Cell Count in Case 2

Group	Cell type	Fraction	Reference range (%)
Cells of myelopoiesis	Myeloblasts	5.0%	0.2-2.9
	Promyelocytes	4.4%	1.5-8.4
	Myelocytes	3.2%	1.0-9.7
	Metamyelocytes	3.8%	3.6-14.6
	Band cells	4.0%	10.6-24.6
	Segmented neutrophils	2.4%	8.5-33.2
	Eosinophils	0.2%	1.2-5.3
	Basophils	0.8%	0-0.8
	Total	23.8%	50.4-70.5
Cells of erythropoiesis	Proerythroblasts	1.4%	0.2-1.3
	Basophilic erythroblasts	2.2%	0.8-6.7
	Polychromatic erythroblasts	61.8%	4.1-29.1
	Orthochromatic erythroblast	2.6%	0.1-5.7
	Total	68.0%	18.4-33.8
Other cell types	Megakaryocytes	0%	0-0.1
	Plasma cells	0.2%	0.2-1.7
	Reticular cells	0.2%	0-3.9
	Lymphocytes	5.8%	5.0-32.6
	Monocytes	2.0%	0.7-6.0

3,100/ μ L with a differential count of 4.0% myeloblasts; and platelet count, 3.0×10^4 / μ L (Figure C). The elevated levels of total bilirubin (2.3 mg/dL) and LDH (458 IU/L) were consistent with a diagnosis of hemolysis. The levels of AST, ALT and γ -GTP were normal, as were the levels of vitamin B12 and folic acid levels. The level of haptoglobin was not measured. Furthermore, BM aspiration revealed 68.0% erythroblasts, 5.0% myeloblasts with 3.03×10^5 / μ L nucleated cells and 50/ μ L megakaryocytes (normal range: 50-150/ μ L) (Figure D, E). As the percentage of erythroblasts was 68.0%, the percentage of non-erythroid cells was 32.0%. Therefore, the proportion of 5.0% of the myeloblasts in 32.0% of the non-erythroid cells was 15.6%. The erythroblasts exhibited megaloblastic-like abnormalities with lobulated nuclei. The neutrophils demonstrated degradation, and the number of mature neutrophils was decreased. The number of megakaryocytes was not increased; however, micro-megakaryocytes were observed (Table 2). The proportion of myeloblasts was greater than 20% of non-erythroid cells in the BM biopsy specimen. Immunostaining of the BM biopsy specimen was negative for CD34, positive for glycophorin C and partially positive for fetal Hb (HbF). The patient was diagnosed with AEL. A cytogenetic analysis of 20 cells demonstrated 48, XY, +8, +21 in all cells. Subsequently, the patient was referred to another hospital for high-dose chemotherapy with BM transplantation.

Discussion

AEL (M6) is a rare type of leukemia, accounting for less than 5% of all acute myeloid leukemia (AML) cases. In the 1985 revision of the French-American-British (FAB) classification (15), the AML-M6 diagnostic criteria were refined to include the following: erythroblasts represent $\geq 50\%$ of all nucleated BM cells; prominent dyserythropoiesis; and myeloblasts represent $\geq 30\%$ of non-erythroid BM cells. Consequently, the two types of AEL recognized include the

erythroid/myeloid type and the "pure" type. In erythroid/myeloid AEL, erythroid precursors represent $>50\%$ of all nucleated BM cells and myeloblasts account for $>20\%$ of all non-erythroid cells (16-21). Both cases 1 and 2 were of the erythroid/myeloid AEL type.

The clinical features are often relatively nonspecific and include pallor, fever and hepatosplenomegaly. Extramedullary presentations, such as myeloid sarcoma, are unusual in patients with AEL; however, extramedullary sites can be involved, and the diagnosis of AEL is usually established based on the findings of lymph node biopsies. Anemia is often severe in patients with AEL, with a mean hemoglobin level of 7.5 g/dL reported (22). Domingo-Claros A et al. reviewed a series of 62 patients with erythroid neoplastic proliferation. Fifty-three patients met the criteria for the M6a subtype of the WHO classification, and two patients were classified as having pure erythremia (M6b). Seven of the cases could not be classified according to the WHO criteria in 2001. The median MCV was 96 fL (range: 71-118) (23). As the MCV in both of the present cases was greater than 120 fL, the patients exhibited markedly high MCVs, even for patients with AEL. Thrombocytopenia and leukopenia, to varying degrees, are also common. Up to one-third of patients can develop hemorrhage, hepatomegaly or splenomegaly. In general, AEL exhibits an aggressive clinical course. Less favorable outcomes are observed in the elderly, patients with a previous diagnosis of MDS and those who have undergone chemotherapy for previous neoplasms (22). Therefore, high-dose chemotherapy followed by BM transplantation or peripheral blood stem cell transplantation is recommended for all patients with AEL (24).

Both of the current patients presented with macrocytic anemia, with peripheral reticulocytosis contributing to at least some extent (6% of peripheral RBCs in case 1 and 24% of peripheral RBCs in case 2). Because a normal reticulocyte MCV is only 103-126 fL (8-11), reticulocytosis was unlikely to be the only factor contributing to macrocy-

tosis in these cases, as the MCV was >120 fL in both patients, and the reticulocyte counts were considerably <100%. Therefore, some other factors, such as MDS, may have also accounted for the macrocytic anemia observed in these patients. The karyotype of 48, XY, +8, +21 demonstrated in case 2 is sometimes seen in patients with MDS. However, even if the coexistence of MDS was present, MDS alone could not have caused macrocytic anemia with an MCV >120 fL. Therefore, marked reticulocytosis and underlying MDS or erythropoiesis abnormalities due to AEL additively could have caused the marked macrocytic anemia. Neither of the patients were alcohol abusers, and there were no findings suggestive of liver disease.

The present patients exhibited the characteristics of hemolytic anemia, including elevated total bilirubin and LDH levels and decreased haptoglobin levels. Hemolysis can occur in patients with AEL and may be severe (25). Although we do not know whether the hemolysis observed in these cases was responsible for the severe macrocytosis, it is likely that the abnormal RBCs produced by AEL-affected BM are more easily destroyed, thereby accounting for the hemolysis.

The number of reported cases of AEL is quite small compared with the more typical causes of macrocytic anemia, such as folate or cobalamin deficiency, liver disease, including alcoholism, and adverse drug effects, as well as other causes of hemolysis. However, the details of the present cases suggest that AEL should be considered in the differential diagnosis of macrocytic anemia, especially when hemolysis and/or peripheral reticulocytosis are present.

The authors state that they have no Conflict of Interest (COI).

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