

Pulmonary Thromboembolism Associated With Mixed-Type Autoimmune Hemolytic Anemia

Mingfeng Lee^a, Takakazu Higuchi^{b, c}, Ryosuke Koyamada^b, Sadamu Okada^b

Abstract

A 56-year-old woman presented with mixed-type autoimmune hemolytic anemia (AIHA) and was complicated with deep vein thrombosis (DVT) and pulmonary thromboembolism (PTE). While DVT/PTE has been recently recognized as a complication of AIHA, there has been no reported case of mixed-type AIHA complicated with DVT/PTE. Mixed-type AIHA is a rare subtype of AIHA and both warm- and cold-type autoantibodies were assumed to have contributed to the development of VTE/PTE in the present case. As severe AIHA and PTE share common symptoms, it is important to consider a possibility of the complication of PTE in the management of AIHA patients.

Keywords: Autoimmune hemolytic anemia; Mixed-type; Deep vein thrombosis; Pulmonary thromboembolism; Complements

Introduction

Autoimmune hemolytic anemia (AIHA) is a rare autoimmune disorder in which autoantibodies against red blood cell (RBC) surface antigens are produced, resulting in premature peripheral destruction of RBCs, with estimated incidence in adults being 1 - 3 per 100,000 person-years [1, 2].

The majority of AIHA have either warm-type AIHA, cold agglutinin disease (CAD), or paroxysmal cold hemoglobinuria (PCH); however, some AIHA patients simultaneously develop both warm- and cold-type autoantibodies and are referred to as mixed-type AIHA [3-5]. On the other hand, while it is still not widely recognized that AIHA is a risk factor of deep vein thrombosis (DVT)/pulmonary thromboembolism (PTE), 10-25% of AIHA patients are reported to be complicated with

thromboembolism and a considerable number of AIHA patients complicated with DVT/PTE are unrecognized and, thus, not treated [6-10]. Moreover, to our knowledge, complication of thromboembolism in patients with mixed-type AIHA has not been reported before and the contribution of each anti-RBC autoantibody subtype to the development of thromboembolism is unknown.

We herein report a case of mixed-type AIHA complicated with DVT/PTE at presentation of AIHA.

Case Report

A 56-year-old Japanese woman was admitted for the evaluation and treatment of severe anemia. She had been suffering from progressive weakness, dizziness, and dyspnea which had started 2 weeks before. She visited another hospital and severe anemia with jaundice was noted. The hemoglobin (Hb) level was 2.8 g/dL, the total bilirubin (T-Bil) was 6.0 mg/dL, and the lactate dehydrogenase (LDH) was 963 U/L. Packed RBCs were transfused and, on the next day, she was transferred to our hospital. She had no past medical history except depression for which she had been taking fluvoxamine, mianserin and etizolam for 8 years. She had quit smoking 3 years before. She had a daughter and had no family member who had any kind of hematological or autoimmune diseases.

On transfer, she was pallor and the conjunctivae were icteric. The liver and the spleen were not palpable. Neither subcutaneous nor mucosal hemorrhage was noted. The laboratory data showed that the white cell blood count was $6.3 \times 10^9/L$, the RBC count was $1.38 \times 10^{12}/L$ with 13.14% reticulocytes, the Hb level was 6.1 g/dL, the hematocrit was 15.1%, and the platelet count was $82 \times 10^9/L$. The coagulation tests revealed that the D-dimer was elevated to 62.7 $\mu g/dL$. The blood chemistry showed that the T-Bil was 6.0 mg/dL, the aspartate aminotransferase was 42 U/L, the alanine aminotransferase was 16 U/L, and the LDH was 974 U/L. The haptoglobin level was below the level of detection. The direct and indirect Coombs' tests were both positive and monospecific antibodies revealed that the RBCs were coated with immunoglobulin (Ig) G and fragments of the third component of complement (C3). The IgG level was 914 mg/dL, IgA was 280 mg/dL, IgM was 89 mg/dL, C3 was 43 mg/dL (reference range, 65 - 135) and the fourth component of complement (C4) was 2 mg/dL (refer-

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^aDepartment of Internal Medicine, St. Luke's International Hospital, Tokyo, Japan

^bDivision of Hematology, St. Luke's International Hospital, Tokyo, Japan

^cCorresponding Author: Takakazu Higuchi, St. Luke's International Hospital, 1-9, Akashi-cho, Chuo-ku, Tokyo 104-8560, Japan. Email: takhig@luke.ac.jp

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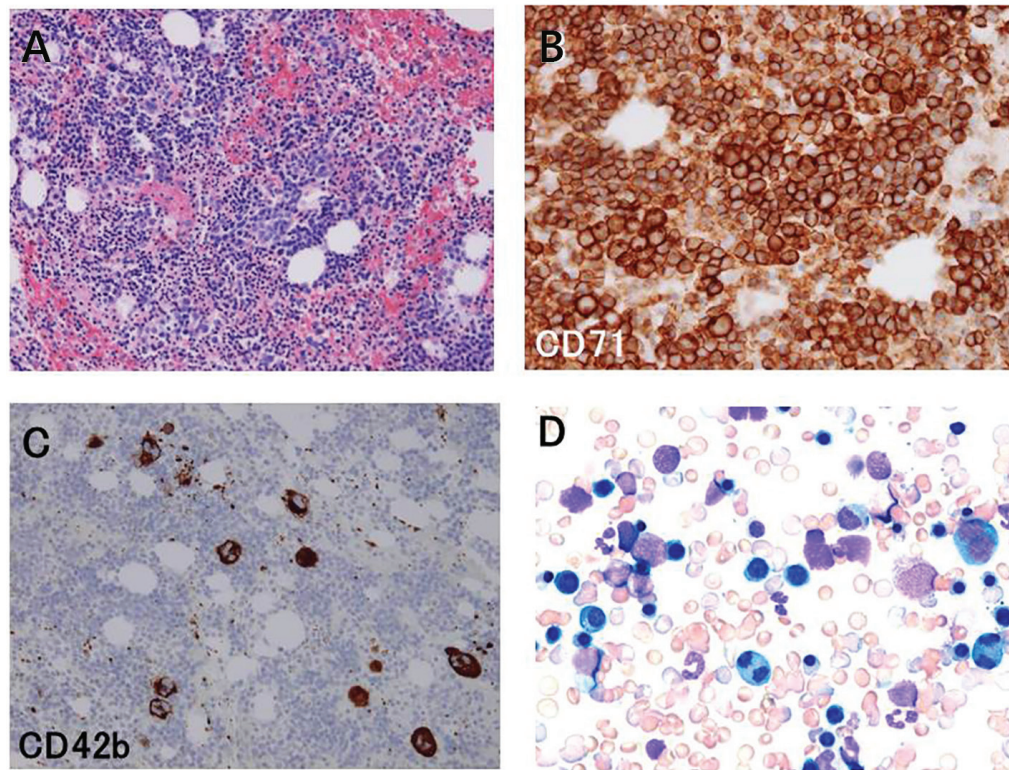


Figure 1. Examination of clot section specimen (A, B, and C) and wedge smear cytology (D) of the bone marrow aspirate. Hematoxylin and eosin stain (A) showed hypercellular bone marrow ($\times 100$). Immunostaining with antibodies against CD71 (B) and CD42b (C) revealed that the erythroid cells were markedly increased and the megakaryocytes were also increased ($\times 200$). Immature and binucleated erythroid cells were frequently observed (D, $\times 200$).

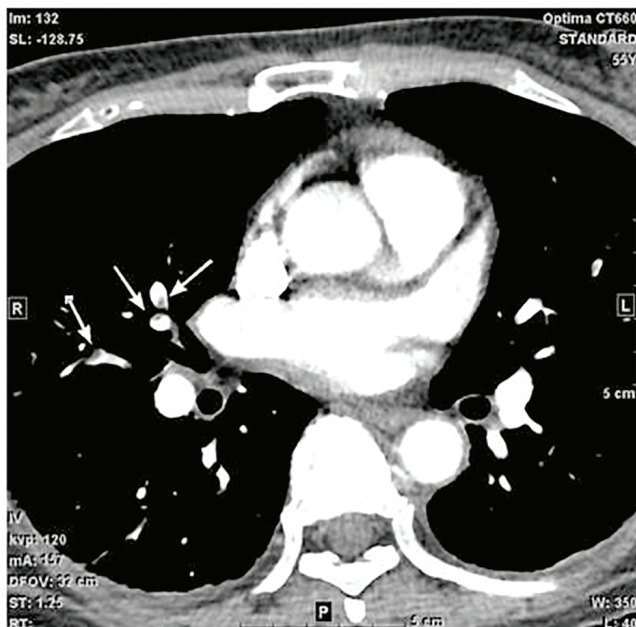


Figure 2. Contrast-enhanced computed tomography (CT) of the chest showed multiple filling defects in the bilateral pulmonary arteries (arrows). These findings are diagnostic of multiple acute pulmonary thromboembolism.

ence range, 13 - 35). The platelet-associated (PA) IgG was 73.2 ng/ 10^7 cells (reference range, 5.0 - 25.0). The antinuclear antibody was positive with a ratio of 1:80 with homogenous pattern. Other autoantibodies studied, including anti-double stranded DNA, anti-smith, anti-SS-A, anti-cardiolipin, and anti-cardiolipin- $\beta 2$ glycoprotein I antibodies, and lupus anticoagulant were all negative. The cold agglutinin titer was 1:256. The hepatitis B surface antigen, anti-hepatitis C virus, anti-human immunodeficiency virus, and anti-adult T-cell leukemia virus antibodies, and anti-*Helicobacter pylori* IgG were negative. The bone marrow examination showed marked erythroid hyperplasia with increased megakaryocytes (Fig. 1). Warm-type AIHA possibly with immune thrombocytopenia was diagnosed and oral prednisolone (PSL) was started at 1 mg/kg/day with RBC transfusion to maintain the Hb level above 7.0 g/dL.

Although she did not have chest symptoms, the elevated D-dimer level and dyspnea suggested PTE and contrast-enhanced computed tomography was performed, which showed multiple filling defects in the bilateral pulmonary arteries diagnostic of multiple acute PTE (Fig. 2). In addition, DVT of the right popliteal vein was detected. The anticoagulation therapy with heparin was started.

The thermal amplitude of the cold agglutinin was studied by incubating O type RBCs and the serum of the patient, which was serially diluted in normal saline, at room temperature, 4,

Table 1. Thermal Amplitude of the Cold Agglutinin

Dilution	× 1	× 2	× 4	× 8	× 16
4 °C	2+	2+	1+	Weak	0
RT	1+	Weak	0	0	0
30 °C	1+	1+	0	0	0
37 °C	Weak	0	0	0	0

The patient's serum was serially diluted in normal saline and incubated with O type RBCs at room temperature, 4, 30, and 37 °C for 45 min and agglutination was determined. RT: room temperature.

30, and 37 °C (Table 1). The patient's serum contained cold agglutinins which were active at 30 °C and judged to have wide thermal amplitude and the patient was diagnosed as mixed-type AIHA.

The anemia responded to PSL and the platelet count also increased to $180 \times 10^9/L$. The Hb level was maintained at between 9 and 10 g/dL with a tapered dose of PSL and remained thereafter with low complement levels. With resolution of thrombi, heparin was switched to warfarin and she was discharged.

Discussion

The majority of AIHA cases have either warm-type AIHA, CAD, or PCH based on the characteristics of the causative autoantibody and warm-type AIHA comprises about 75% of all AIHA cases. On the other hand, approximately 6-8% of AIHA patients simultaneously develop both warm-type antibody and cold agglutinin and are referred to as mixed-type AIHA [3-5], although the frequency is reported to be lower when more strict diagnostic criterion is applied [11].

While it is still not widely recognized, DVT/PTE is a rather common complication of AIHA [6-10, 12-16]. The association between DVT/PTE and AIHA has long been recognized since 1967, when Allgood and Chaplin reported that PE was complicated in five of their cohort of 47 patients and was the most common cause of death [17]. More recently, a cross-sectional [13] and three cohort studies [12, 14, 15] reported that the risk of complicating DVT/PTE was increased in AIHA patients and a meta-analysis of these studies demonstrated that the pooled risk ratio of DVT/PTE was 2.63 [16]. While case series of AIHA showed that 10-25% of AIHA patients were complicated with thromboembolism and a substantial number of them had anti-phospholipid antibodies (APLA) [6-10], about 15% of AIHA patients who had thromboembolism did not have APLA, suggesting that AIHA itself is associated with thromboembolism [6, 10]. Various mechanisms have been proposed for the hypercoagulability in AIHA patients. Disruption and loss of RBC membrane expose negatively-charged membrane phosphatidylserine on the surface which provides a surface for formation of the tenase and prothrombinase complex [6]. Antibody-mediated decrease in cytoskeleton protein may also contribute to this process. Cytokine-induced expression of tissue factor on monocytes and endothelium may also be involved [6]. In addition, microparticles released from RBC

fragments during hemolysis are thought to trigger the coagulation cascade [18]. Moreover, sequestration of nitric oxide by free plasma Hb released from damaged RBCs can lead to uninhibited platelet aggregation and vascular smooth muscle dystonia, resulting in clot formation [19]. Notably, as the present case had mixed-type AIHA, the contribution of cold agglutinin to the development of thromboembolism should be considered. Destruction of RBCs in patients with warm-type AIHA is primarily extravascular sequestration by phagocytosis in the spleen and intravascular hemolysis is unusual [20]. In the case of CAD, although the major mechanism of hemolysis is extravascular hemolysis by the reticuloendothelial cells in the liver [21, 22], intravascular hemolysis mediated by terminal complement complex may occur in up to 10% of the patients [23]. Profound anemia and low complement levels seen in the present case suggested that intravascular hemolysis due to the activation of terminal complement complex was operative, releasing excess free Hb into the blood stream, thus contributing to the development of thromboembolism. Presently, the patient is still mildly anemic with low complement levels, which suggests continuous activation of complement system and low grade hemolysis, without any signs of the recurrence of thromboembolism; however, as she is still on anti-coagulation therapy with warfarin, the contribution of cold agglutinin to the development of thromboembolism remains speculative until more cases are studied.

The patient had mild thrombocytopenia and increased megakaryocytes in the bone marrow with elevated PAIgG level and the platelet count subsequently elevated with the start of PSL. These findings suggested a possibility of immune thrombocytopenia and it appeared likely that she had Evans' syndrome. Immune thrombocytopenia has also been reported to be a risk factor of DVT [13, 14]. However, as the consumption of platelet by thrombus formation may have contributed to the decrease in the platelet count, the diagnosis remained unconfirmed.

In conclusion, the present case shows that it is important to consider a possibility of DVT/PTE when treating patients with AIHA because the clinical presentation of severe AIHA may resemble that of PTE, which can be fatal and require immediate optimal intervention.

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