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Cyclic Thrombocytopenia in a Patient Treated With Cyclosporine for Refractory Idiopathic Thrombocytopenic Purpura

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Cyclic thrombocytopenia (CT) is a rare disorder with cyclic changes of the platelet counts. Though the pathogenesis of this disorder has not been clarified, recent reports suggest that periodic destruction and/or ineffective production of platelets may be important causes of the disease. We report a case of a patient with refractory idiopathic thrombocytopenic purpura (ITP) in whom CT developed after cyclosporine A (CyA) therapy. There was an inverse relation between platelet counts and the serum levels of platelet-associated immunoglobulin G (PAIgG). The ploidy of bone marrow megakaryocytes also had an inverse relation with platelet counts. When the platelet count was low, the ploidy of megakaryocytes increased ($P < 0.01$). The number and area of bone marrow megakaryocytes were unrelated to platelet counts. These results indicate the possibility of platelet destruction caused by immunological mechanisms in CT. Cyclosporine A could have certain but fluctuating regulatory effects against antibody production for circulating platelets, which could lead to cyclic changes of the platelet counts. This case also suggests that CyA can be effective in severe refractory ITP. Regulatory mechanisms of platelet production and destruction and appropriate doses of CyA should be further studied in autoimmune-mediated thrombocytopenias. Am. J. Hematol. 56:272–276, 1997.

Key words: cyclic thrombocytopenia; cyclosporine A; idiopathic thrombocytopenic purpura

INTRODUCTION

Idiopathic thrombocytopenic purpura (ITP) is a hemorrhagic disease caused by autoimmune disorders characterized by thrombocytopenia, normal bone marrow, and the absence of other apparent causes of low platelet count [1,2]. Patients who have symptomatic moderate or severe thrombocytopenia are conventionally treated with prednisolone. Splenectomy is also well-recognized therapy and is usually required for patients unresponsive to prednisolone [1,2]. Patients who are refractory to corticosteroids and splenectomy and do not maintain safe platelet counts should be treated with other agents.

Cyclosporine (CyA) is thought to block T-lymphocyte mediated responses and therefore has been given for the purpose of its immunosuppressive effects in organ transplantation recipients [3,4]. Recently, it has become known that CyA is effective for aplastic anemia, pure red cell aplasia, and other autoimmune-mediated hematological disorders [5]. We report a case of refractory ITP that developed cyclic thrombocytopenia (CT) after CyA treatment. In the thrombocytopenic and thrombocytotic cycles, the number, ploidy, and area of bone marrow megakaryocytes were measured. Serum concentration of platelet associated immunoglobulin G (PAIgG) was also examined at various points throughout the course.

CASE REPORT

A 21-year-old Japanese woman visited Tokyo Metropolitan Fuchu Hospital because of purpura in April 1987.

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TABLE I. The Relationship Between Platelet Counts and Serum PAIgG Number, Ploidy, and Area of Megakarycocytes.

	Date		
	October 28, 1994	November 4, 1994	
Platelet count ($x10^9$/L)	73	16	
PA IgG (ng/10^7cells)	59.7	245.3	
Number of megakaryocytes (/mm^2)	56.9	37.0	
Ploidy of megakaryocytes (N)	16.07 ± 8.65	20.96 ± 7.73	$P < 0.01$
Area of megakaryocytes (μm^2)	257.6 ± 95.7	339.7 ± 156.4	NS

ITP was diagnosed and 60 mg of prednisolone and 100 mg of azathioprine were given consecutively. Elevation of platelet counts was not observed and she was introduced to our hospital for splenectomy in July 1990. Splenectomy on October 9, 1990, was followed by an increase of platelets counts from 9×10^9/L to 30×10^9/L. After discharge in November 1990, she received oral administration of 20 mg/day prednisolone. In January 1991, the platelet count began to decrease, reaching 10×10^9/L. After admission she received methylprednisolone (mPSL) pulse therapy (20 mg/kg/day ×5 days, then tapered). Danazol, azathioprine, and interferon-α were administered without success. Only slow infusions of vincristine sulfate induced a rise in platelet count to 72×10^9/L. She was discharged after several injections of vincristine sulfate but was readmitted in December 1991 with decreased platelet count. On admission, purpura and ecchymosis were remarkable. The platelet count was 5×10^9/L. The white blood cell count (WBC) was 10.9×10^9/L. The hemoglobin concentration was 149 g/L and the red blood cell count was 5.16×10^{12}/L. PAIgG was 616.7 ng/10^7 platelets (normal <25 ng). Anti-nucleic antibody, anti-DNA antibody, LE factor, and anti SS-A antibody were negative and serum levels of C3 and C4 were normal. Bone marrow aspirates revealed normocellularity and megakaryocytes were normal in number. Weekly oral administration of 10 mg of methotrexate was ineffective and mPSL pulse therapy had a temporary effect with an increase of platelet counts up to 330×10^9/L. Then slow injections of vincristine sulfate, vindesine sulfate, and oral administration of colchicine, danazol, and mizoribine were attempted, but effective elevation of platelet counts was not observed. From October 27, 1992, 50 mg/body of CyA was administered and the doses were increased to 200 mg within several days. Combination of CyA and mPSL pulse therapy yielded undulating effects. After a pulse therapy with daily administration of 200 mg of CyA, the platelet count was markedly elevated but fluctuated. The platelet counts ranged from a minimum level of 1×10^9/L to a maximum level of 177×10^9/L. Serum trough levels of CyA were 130 and 150 ng/ml when she had been taking 200 mg/body of CyA daily. Her symptoms gradually ameliorated and she was discharged at the end of February 1993. After discharge, her platelet count again decreased to 1×10^9/L, though she had continued taking 200 mg CyA. She was again admitted and mPSL pulse therapy with administration of CyA was begun in May 1993. The platelet count was elevated to 130×10^9/L and 298×10^9/L on 22 and 34 days after the pulse therapy. On July 2, 1993, her platelet count reached its peak of 619×10^9/L, but the maximum values of the platelet count again fluctuated, with a minimum level of 1×10^9/L. mPSL pulse therapy was halted and the platelet count became cyclic with a period of from 15 to 30 days. The hemorrhagic diathesis improved after the platelet counts became cyclic and the patient was discharged at the end of July. In early October 1993, she refused CyA, then her platelet level began to decrease and genital bleeding occurred. In December she began receiving 125 mg of CyA and the platelet counts again became cyclic, though the maximum value was lower than before. She has continued taking 100–125 mg CyA daily. The platelet counts are still cyclic, ranging from 1×10^9/L to 581×10^9/L and symptoms have improved without further additional treatment (Fig. 1).

There was an inverse relation between the platelet count and the serum concentration of PAIgG. When the platelet count was less than 5×10^9/L, the serum PAIgG was usually over 100 ng/10^7 platelets. Its highest value was 724.5 ng/10^7 platelets, when the platelet count was 1×10^9/L. On the other hand, the lowest value was 7.6 ng/10^7 platelets when the platelet count was 380×10^9/L (Fig. 2). This inverse relation was not influenced by treatment throughout the entire course.

The number, ploidy, and area of bone marrow megakaryocytes were measured twice during administration of CyA, once when the platelet count was 73×10^9/L and also when it was 16×10^9/L (Table I). The number of megakaryocytes was counted light microscopically in a clot section of aspirated bone marrow that was stained by May-Grünwald-Giemsa stain solution. The ploidy and area of megakaryocytes were measured as follows: bone marrow smear was stained by Feulgen solution and the amounts of DNA of 50 megakaryocytes were measured with Quantitative Ploidy Analysis and Cell Measurement Program (CAS, Cell Analysis Systems, Inc, Elmhurst, IL). The mean DNA amounts of 10 mature neutrophils in the same smear were designated as 2N and were compared with those of megakaryocytes. The area of megakaryocytes was also measured with the CAS. Bone marrow aspirates were stained with May-Grünwald-Giemsa stain solution and the area of 50 megakaryocytes was measured with the CAS. No significant difference was observed in the megakaryocyte count and area, but there

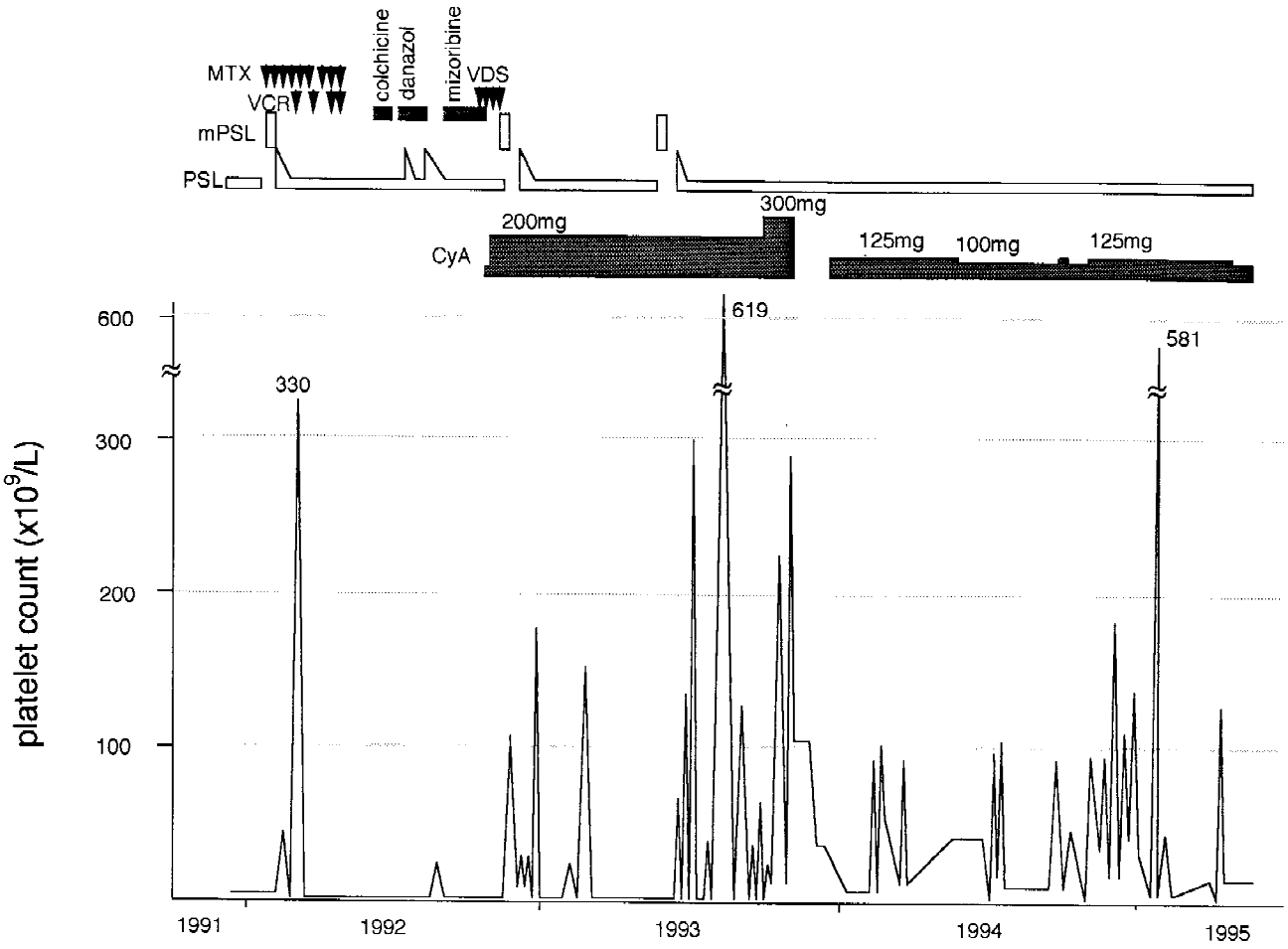

Fig. 1. Cyclic evolution of platelet counts after cyclosporine A administration. CyA: cyclosporine A; PSL: prednisolone; mPSL: methylprednisolone; MTX: methotrexate; VCR: vincristine sulfate; VDS: vindesine sulfate.

was a statistical difference in the ploidy of megakaryocytes (16.07 ± 8.65 vs. 20.96 ± 7.73, $P < 0.01$).

DISCUSSION

Cyclic thrombocytopenia is a rare disorder with cyclic changes of platelet levels ranging from several thousand (thrombocytopenia) to as high as several hundred thousand/μl [6]. The pathogenesis of this disorder remains unclear. In women, platelet counts physiologically fluctuate with the menstrual cycle [7], but the fact that CT has occurred in men and also in women after menopause indicates that the pathogenesis of this disorder is not necessarily related to the menstrual cycle. In our patient there was no relationship between the platelet cycles and menstruation.

Several reports have suggested that platelet destruction and/or periodic failure of effective platelet production may be important in the pathogenesis of this disease [8–12].

In the present case, there was an inverse relation between platelet counts and serum level of PAIgG. More than 90% of PAIgG content is in alpha-granules in platelets and is released quickly on stimulation by ADP and thrombin. Greatly increased PAIgG examined within 5 to 6 hr after harvesting blood indicates a high concentration of surface PAIgG, which may represent antiplatelet antibody in patients with ITP [13]. To avoid measuring intracellular PAIgG, blood specimens were examined as soon as possible. A high value of PAIgG had been observed before splenectomy when the platelet count had been continuously low. An inverse relation between PAIgG and platelet levels in CT patients has been reported by some investigators [9,11]. Their study suggested cyclic production of autoantibodies to the platelet membrane glycoprotein (GP) II b/III a complex to be of pathologic significance in some patients with CT [9,11]. The existence of autoantibodies to GP II b/III a has been well documented in ITP [14]. Recently, it has been reported that platelet-associated IgM anti GP II b/III a au-

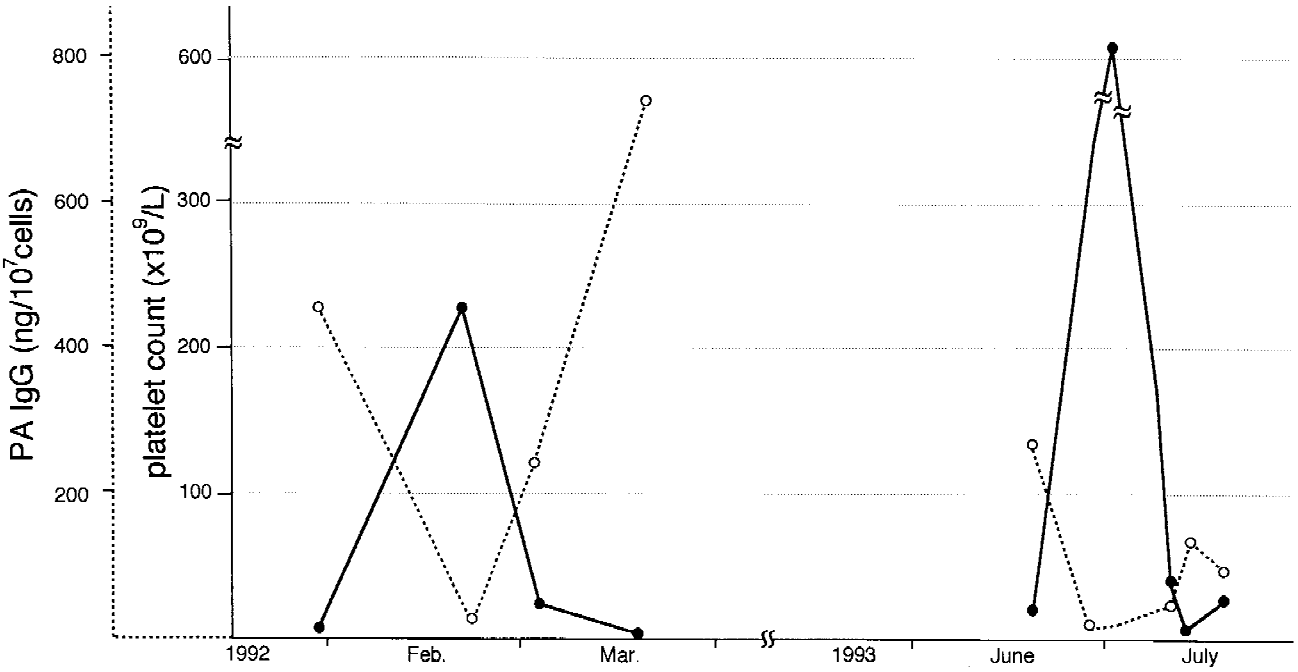

Fig. 2. The inverse relationship between serum PAIgG (broken line) and platelet counts (solid line) during two cycles.

toantibodies are also associated with CT [12]. In our patient, the platelet count had a tendency to increase along with the decrease of PAIgG after CyA had been given. This fact may suggest the possibility of immunological destruction of the circulating platelets, which might be partially affected by CyA.

Differences in the number and area of bone marrow megakaryocytes showed no statistical relationship with differences in the platelet counts on the days on which bone marrow aspiration was performed. The ploidy of megakaryocytes was statistically different at the two points in the platelet cycle. When the platelet count increased, the ploidy of megakaryocytes was lower, whereas it was higher when the platelet count decreased. It is not certain whether this indicates the etiological background of CT. Higher ploidy could be a result of feedback mechanisms that recognize fewer platelet counts in the peripheral blood. The cyclic nature of the platelet count while on CyA could be related to variations in survival, production, or both. To fully elucidate this point, a method for the accurate and reliable measurement of platelet survival would be necessary. At present, however, there is no generally established method to evaluate platelet survival, although labelling with radioisotope, including Indium, may be one possibility, if platelet counts are sufficiently high. Future studies on this topic should seek to determine platelet survival in order to clarify the roles of survival and production in this phenomenon.

Kelsey et al. reported the usefulness of CyA in severe refractory ITP [15]. In their patient, and in our case, no treatment except vincristine was reliably effective. CyA increased the platelet count to 100×10^9/L for over 2 months in their patient. Schultz et al. administered CyA in several cases of childhood ITP [16]. They reported that CyA had merely a temporary and minor effect. They indicated a relation between serum levels of CyA and increase of platelet counts and showed that relatively high levels of CyA (>500 ng/mL) were required to obtain increases of platelet count over 50×10^9/L. As renal toxicity is a major side effect of this drug and is augmented in a dose-dependent manner [17], we hesitated to increase the doses and had kept serum levels below 200 ng/ml. Velu et al. reported the first case of refractory ITP developing CT after CyA treatment [18]. In their case, doses of CyA were 6–12 mg/kg and the platelet count fluctuated from 1 to 744×10^9/L. PAIgG ranged from 350–750 ng/10^7 platelets. The relation between platelet count and PAIgG was not shown nor was the serum level of CyA mentioned. The dosage of CyA we selected was 2–5.5 mg/kg, which was lower than that usually used in autoimmune diseases [17], but it is uncertain whether a greater amount of the drug would improve the prognosis of the patient. While CyA may be useful in severe ITP, an appropriate protocol of CyA in autoimmune thrombocytopenias should be established based on studies of pharmacokinetics and toxicity of the drug.

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