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CASE REPORT

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Pancytopenia, including macrocytic anemia, associated with leflunomide in a rheumatoid arthritis patient

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Abstract A female rheumatoid arthritis patient was admitted for productive cough and general fatigue that had gradually developed after leflunomide therapy. Side effects including severe hypoxia, thrombocytopenia, lymphocytopenia, and macrocytic anemia with schistocytes (probably drug-induced megaloblastic anemia) were noted. Leflunomide-eliminating cholestyramine therapy successfully treated all conditions excluding severe hypoxia, which occurred owing to deteriorating interstitial pneumonia and complicated bacterial pneumonia following antibiotic treatment. This is a rare case of leflunomide-associated multiple hematopoietic impairments.

Key words Drug-induced megaloblastic anemia · Leflunomide · Macrocytic anemia · Pancytopenia

Introduction

Leflunomide (Arava; Sanofi-Aventis, Paris, France), an inhibitor of pyrimidine synthesis, is a new disease-modifying anti-rheumatic drug (DMARD). Recently, the use of leflunomide was approved in Japan; it was expected to be a new DMARD that was as effective as methotrexate (MTX) in the treatment of rheumatoid arthritis (RA). The mechanism of action of leflunomide is considered to involve inhibition of dihydroorotate dehydrogenase (DHODH), a key enzyme involved in the biosynthesis of pyrimidine nucleotide triphosphates. Leflunomide was more advantageous than MTX in that it aggravated interstitial pneumonia (IP) to a relatively less extent. However, a post-marketing survey revealed reports on the deterioration of conditions present in a patient orally administered with leflunomide; subsequently, this drug was contraindicated in patients with a previous history of IP.[1–7] However, other severe leflunomide-related side effects, including pancytopenia, have also been reported.[8,9] Our patient developed cough and marked hypoxia after leflunomide therapy. In addition, thrombocytopenia, lymphocytopenia, and macrocytic anemia with schistocytes were detected. We present an interesting case of leflunomide-associated pancytopenia, including macrocytic anemia.

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Case report

On February 13, 2004, a 69-year-old woman complaining of general fatigue and cough was admitted to our hospital. In 1991, she was diagnosed with RA; subsequently, she was followed up in another hospital. In 1997, she sustained a fracture to the left ankle joint because of a traffic accident; she was admitted to the Department of Orthopedics in our hospital and was administered oral prednisolone for the treatment of RA at our department. In 2000, the patient developed fever, and she was subsequently diagnosed with IP followed by treatment with methylprednisolone pulse therapy. In July 2003, the patient developed sudden-onset polyarthralgia, and MTX therapy was started. In October 2003, MTX was substituted with leflunomide because of persistent polyarthralgia. This resulted in an improvement in the condition, and the C-reactive protein (CRP) level decreased to that within the normal range. Nonetheless, there was a gradual onset of productive cough, and she experienced general fatigue. In January 2004, leflunomide therapy was discontinued because of the deterioration of IP; however, there was further progression of cough and general fatigue, and she was admitted to our hospital on February 13, 2004.

Physical examination performed on admission revealed anemic palpebral conjunctiva and audible fine crackles at

Table 1. Laboratory findings on admission

CBC	LDH 637 IU/l	C4 30 mg/dl
WBC 5100 μl^{-1}	LDH isozyme	CH50 47.1 U/ml
Lymphocytes 200 μl^{-1}	1 32%	RF 68.8 IU/ml
RBC $223 \times 10^4 \mu l^{-1}$	2 37%	Vit. B_{12} 750 pg/ml
Hb 7.5 g/dl	3 17%	Folic acid 3.7 ng/ml
Ht 23.7%	4 6%	
MCV 106.3 fl	5 8%	*Urinalysis*
MCH 33.6 pg	TB 1.2 mg/dl	SG 1.014
MCHC 31.6%	DB 0.1 mg/dl	pH 6.0
PLT $8.6 \times 10^4 \mu l^{-1}$	ALP 282 IU/l	Pro (+2)
RDW 20.6%	γ-GT 61 IU/l	Glu (+3)
Anisocytosis (+)	BUN 19 mg/dl	Urobilinogen (±)
Poikilocytosis (+)	Cr 0.6 mg/dl	Acetone (–)
Nucleated red blood cells (+)	Na 144 mmol/l	Bilirubin (–)
Schistocytes (+)	K 3.0 mmol/l	OB (+1)
Macro-ovalocytes (+)	Cl 103 mmol/l	RBC 1 ~ 4/HPF
	CRP 0.91 mg/dl	WBC 1 ~ 4/HPF
Coagulation	Fe 75 μg/dl	
PT 100%	UIBC 77 μg/dl	*ABG (room air)*
APTT 25.2 s	Ferritin 194 ng/ml	pH 7.460
Fbg 421 mg/dl	Haptoglobin 29 mg/dl	$PaCO_2$ 46.8 mmHg
	KL-6 518 U/ml	PaO_2 52.3 mmHg
Blood chemistry and serology	SP-D 99.7 ng/ml	HCO_3 32.8 mmol/l
TP 5.0 g/dl	IgG 457 mg/dl	BE 8.4 mmol/l
Alb 3.0 g/dl	IgA 334 mg/dl	SaO_2 88.4%
AST 12 IU/l	IgM 36 mg/dl	
ALT 11 IU/l	C3 83 mg/dl	

CBC, complete blood cells; WBC, white blood cells; RBC, red blood cells; Hb, hemoglobin; Hct, hematocrit; MCV, mean corpuscular volume; MCH, mean corpuscular hemoglobin; MCHC, mean corpuscular hemoglobin concentration; PLT, platelet; RDW, red cell distribution width; PT, prothrombin time; APTT, activated partial thromboplastin time; Fbg, fibrinogen; TP, total protein; Alb, albumin; AST, aspartate aminotransferase; ALT, alanine aminotransferase; LDH, lactate dehydrogenase; TB, total bilirubin; DB, direct bilirubin; ALP, alkaline phosphatase; γ-GT, γ-glutamyl transpeptidase; BUN, blood urea nitrogen; Cr, creatinine; CRP, C-reactive protein; UIBC, unsaturated iron-binding capacity; SP-D, surfactant-D; Ig, immunoglobulin; RF, rheumatoid factor; Vit, vitamin; SG, specific gravity; Pro, protein; Glu, glucose; OB, occult blood; HPF, high power field; ABG, arterial blood gas; BE, base excess

bilateral lung bases. Laboratory findings were as follows (Table 1): urine protein and occult blood, positive; erythrocyte sedimentation in the urine, negative; decreased lymphocyte count 200 μl^{-1}; hemoglobin level 7.5 g/dl; mean corpuscular volume (MCV) 106.3 fl; increased reticulocyte count $14.3 \times 10^4 \mu l^{-1}$ (on the sixth day after cholestyramine administration); and decreased platelet count $8.6 \times 10^4 \mu l^{-1}$. Additionally, anisocytosis and poikilocytosis were detected in the peripheral blood (Fig. 3A); schistocytes, macro-ovalocytes, and nucleated red blood cells were detected. There was an increase in the level of lactic dehydrogenase (LDH, 637 IU/l; normal range 130–235 IU/l), wherein types 1 and 2 were dominant; that of total bilirubin (TB 1.2 mg/dl; normal range 0.2–1.3 mg/dl) was slightly increased; decreased level of haptoglobin (29 mg/dl; normal range 95–330 mg/dl); and the levels of vitamin B_{12} (750 pg/ml; normal range 233–914 pg/ml) and folic acid (3.7 ng/ml; normal range 2.4–9.8 ng/ml) were within the normal limit; KL-6 level was marginally elevated at 518 U/ml (normal <500 U/ml); β-D glucan level was within normal limits (<4.0 pg/ml; normal <10 pg/ml); and cytomegalovirus antigenemia, negative. Blood gas analysis revealed marked hypoxia. Sputum sample culture revealed the growth of *Pseudomonas aeruginosa* but not of *Pneumocystis jiroveci* or fungi. On admission, chest computed tomography (CT) showed slight ground-glass opacity surrounding the earlier fibrotic change (Fig. 1A).

Because of the deterioration of IP, methylprednisolone semi-pulse therapy followed by oral prednisolone (40 mg) was started on the first day after admission. Subsequently, the productive cough improved slightly; however, hypoxia persisted. On the ninth day, leukocyte (WBC) count increased to 17 400 μl^{-1}; CRP level increased to 3.27 mg/dl; chest CT revealed pleural effusion and bilateral infiltrates in the ground-glass opacities (Fig. 1B). Antibiotic treatment against *P. aeruginosa* was administered immediately to the patient for the treatment of the *P. aeruginosa* infection. Consequently, the hypoxic status of the patient gradually improved; further, the chest CT obtained on the 32nd day did not reveal any aberrant shadow and pleural effusion (Fig. 1C).

For the treatment of lymphocytopenia and thrombocytopenia, drug elimination by cholestyramine was started immediately; cholestyramine administration resulted in immediate recovery from these above-mentioned conditions (Fig. 2).

In addition, macrocytic anemia with schistocytes was detected on admission. However, oral administration of cholestyramine resulted in an immediate improvement in the conditions related to anemia and the MCV decreased. Further, anisocytosis and poikilocytosis decreased and then resolved completely (Fig. 3B). On the 20th day, TB and LDH levels decreased to 0.5 mg/dl and 368 IU/l, respectively.

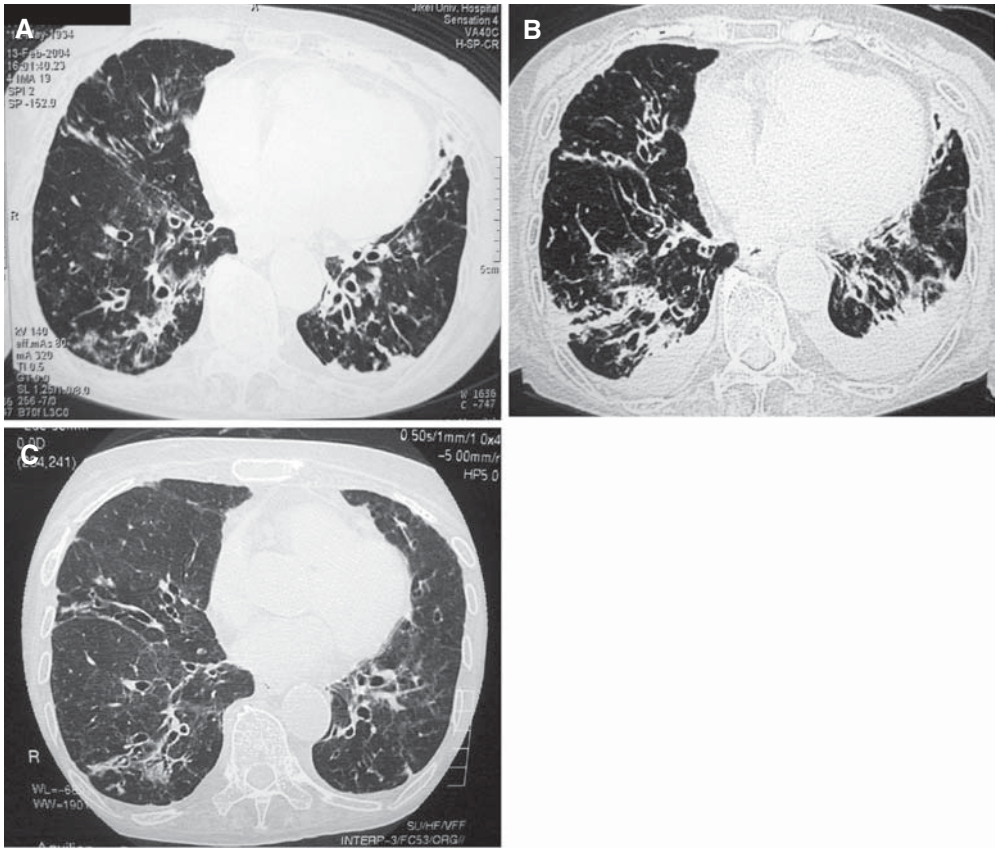

Fig. 1. Computed tomography of chest. **A** On admission (February 13, 2004). **B** The ninth day (February 21, 2004). **C** The 32nd day (March 15, 2004). On admission, mild ground-glass opacities surrounding the previous fibrotic changes were observed (**A**). Subsequently, bilateral infiltrates on ground-glass opacities and pleural effusion appeared (**B**). After antibiotic administration, bilateral infiltrates and pleural effusion disappeared almost completely on the 32nd day (**C**)

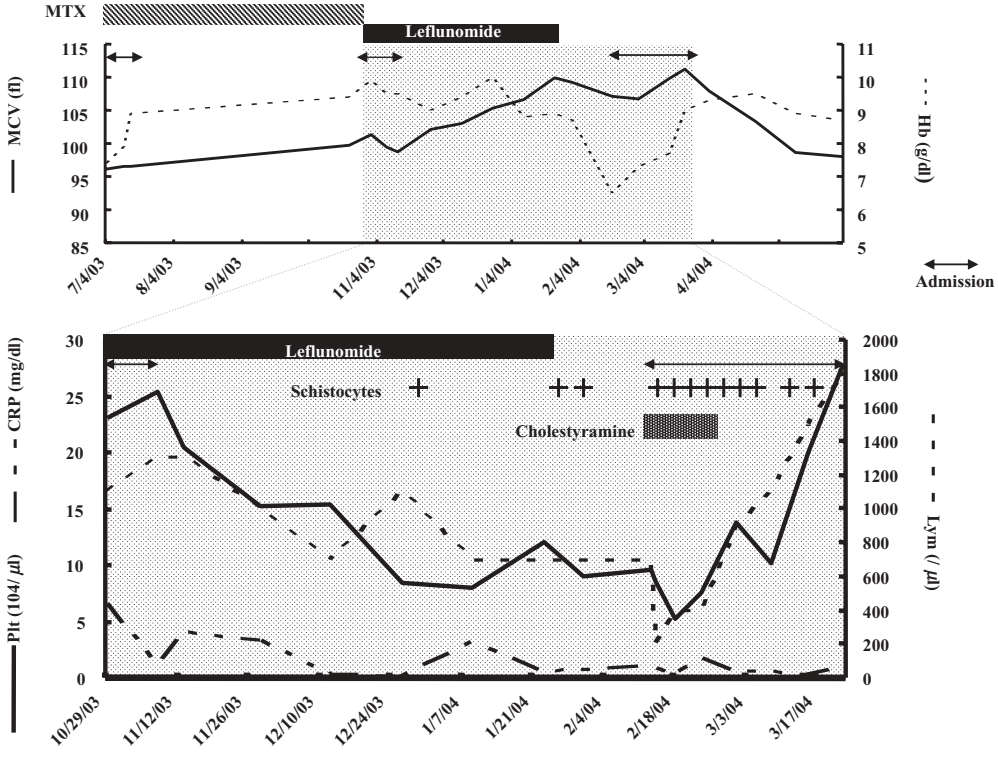

Fig. 2. Time course of macrocytic anemia and clinical course from the time of leflunomide administration until discharge. After substituting methotrexate with leflunomide, there was a simultaneous decrease in the progression of macrocytosis and decreased in the hemoglobin levels. Moreover, thrombocytopenia and lymphocytopenia also developed, and there were positive signs of schistocytes during the last 10 days of December 2003. Cholestyramine administration resulted in a prompt and successful treatment of pancytopenia, including macrocytic anemia

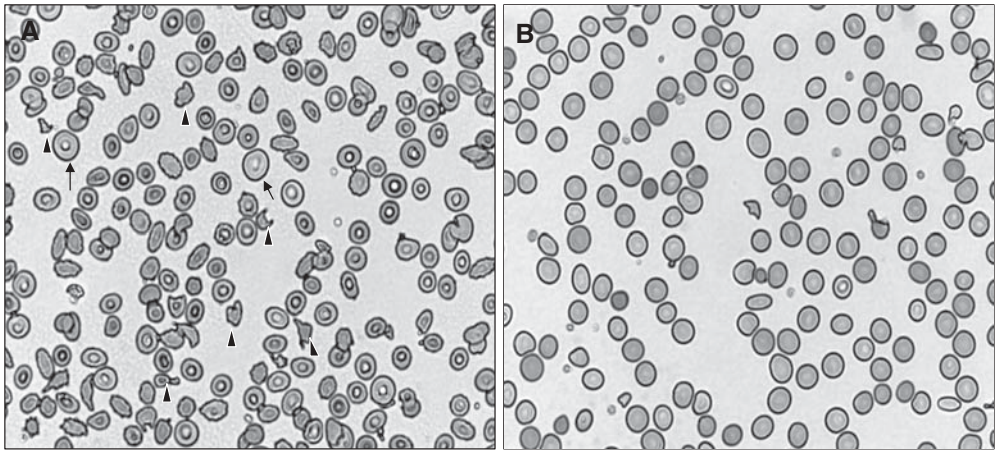

Fig. 3. Transition of the peripheral blood specimen (May-Grünwald Giemsa stain, ×400). **A** On the second day (February 14, 2004). **B** On the 32nd day (March 15, 2004). The findings on the second day revealed anisocytosis, poikilocytosis, macro-ovalocytes (*arrows*), and schistocytes (*arrowheads*) (**A**). However, there was a marked improvement in these conditions after cholestyramine administration (**B**)

Subsequently, the proteinuria that was detected on admission disappeared without any treatment, thereby suggesting that the condition was transient. The patient was discharged on the 39th day of admission. One and a half months after discharge, the schistocytes in the peripheral blood disappeared completely.

Discussion

In the case reported here, approximately 2 months after leflunomide was administered, the patient showed signs of macrocytic anemia with schistocytes and macro-ovalocytes in the peripheral blood. These findings are often observed in cases of severe hematopoietic destruction of the bone marrow and megaloblastic anemia (MA).[8] Savage et al. reported 300 patients with macrocytosis, and the presence of macro-ovalocytes in the peripheral blood had a sensitivity of 100% and a specificity of 81.5% for MA. In our patient, bone marrow examination was not performed because the patient did not provide her consent for the procedure. Therefore, the presence of giant erythroblasts in the bone marrow could not be confirmed. However, considering the mechanism that inhibits DNA synthesis of pyrimidines, the case may have been complicated by drug-induced MA. The inhibitors of pyrimidine DNA synthesis often cause drug-induced MA, similar to that caused by anticancer agents.[9] Currently, in this patient, no other causes of macrocytic anemia, including deficiency of serum vitamin B_{12} and folic acid, liver dysfunction, and alcoholism, were observed. The side effects should be considered when an increase of MCV is observed in the peripheral blood when leflunomide is administered.

Macrocytic anemia with schistocytes and thrombocytopenia both caused by thrombotic microangiopathic hemolytic anemia (TMHA) should also be considered; TMHA occurs in thrombotic thrombocytopenic purpura (TTP) and hemolytic uremic syndrome (HUS). However, anisocytosis and poikilocytosis, which are generally observed in the case of MA, were detected in the peripheral blood. Moreover, the reticulocyte count increased on the sixth day after drug elimination by cholestyramine; reticulocytosis is often observed in the post-treatment stage of MA, whereas a low reticulocyte count is indicative of the pre-treatment stage of MA. TMHA is usually a severe condition and requires the administering of advanced treatment such as plasmapheresis. These findings suggest that MA rather than TMHA was the more probable cause of macrocytic anemia with schistocytes.

In this case, the development of cough and marked hypoxia after substitution of MTX with leflunomide suggested the possibility of deterioration of IP. In our patient, the lymphocyte count and immunoglobulin G level decreased to $200\,\mu l^{-1}$ and 457 mg/dl (normal range 870–1700 mg/dl), respectively, on admission. These findings are often observed in cases of a DMARD-related pulmonary injury.[10,11] However, the sputum sample culture revealed the growth of *P. aeruginosa*. The risk factors of *P. aeruginosa* infection include corticosteroid therapy (>10 mg of prednisolone per day).[12] In our patient, oral prednisolone (15 mg) was administered before admission, and methylprednisolone semi-pulse therapy followed by oral prednisolone (40 mg) was administered after admission. Hence, *P. aeruginosa*-induced pneumonia may have been complicated on the basis of the immunocompromised status of the patient. However, infiltrates are often observed in an air-bronchogram as a result of DMARD-related pulmonary injury.[5] It was unclear whether lung infection only was responsible for the direct etiology of prolonged hypoxia.

It has been reported that leflunomide was associated with pancytopenia.[13,14] Chan et al. suggested that the risk of myelotoxicity associated with leflunomide was increased in older patients (median age, 65.5 years) and on administering a combination of leflunomide and MTX. In the present case, although the patient was elderly, leflunomide and MTX were not administered simultaneously. Regular monitoring is a mandate when leflunomide is administered to older patients. According to the post-marketing survey report of all Japanese patients, until August 23, 2006, 10 of 6079 patients treated with leflunomide had developed pancytopenia (http://safety.sanofi-aventis.co.jp/arava/aravad/, in Japanese). However, the development of macrocytic anemia has not yet been reported. In the present case report,

pancytopenia, including a rare form of anemia, was also detected during leflunomide therapy. To our knowledge, not many similar cases have been reported earlier; therefore, this case is of particular interest.

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