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【 CASE REPORT 】

Acute Kidney Injury by Renal Hemosiderosis Secondary to Primary Cold Agglutinin Disease Associated with an Excessive Alcohol Intake

Aya Imafuku[1], Go Yamamoto[2], Koji Takemura[1], Eiko Hasegawa[1], Naoki Sawa[1], Masahiro Kawada[1], Akinari Sekine[1], Junichi Hoshino[1], Kenmei Takaichi[1], Takeshi Fujii[3], Kenichi Ohashi[4] and Yoshifumi Ubara[1,5]

Abstract:
Renal hemosiderosis occurs in the context of severe intravascular hemolysis, with the most common cause being paroxysmal nocturnal hematuria. Patients with cold agglutinin disease (CAD) have relatively mild hemolysis, and acute kidney injury (AKI) due to renal hemosiderosis has not been reported. We encountered a patient with CAD caused by lymphoplasmacytic lymphoma who developed AKI secondary to renal hemosiderosis after an excessive alcohol intake.

Key words: acute kidney injury, renal hemosiderosis, intravascular hemolysis, cold agglutinin disease, lymphoplasmacytic lymphoma, excessive alcohol intake

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Introduction

Renal hemosiderosis occurs as a consequence of severe intravascular hemolysis. The majority of patients with renal hemosiderosis have paroxysmal nocturnal hematuria (PNH) (1, 2), and only a few cases have been reported due to other causes, such as prosthetic heart valves, ABO-incompatible transfusion, sickle cell anemia, or primary hemochromatosis (3-5).

Primary cold agglutinin disease (CAD) accounts for 15% of autoimmune hemolytic anemia (6), and half of these patients have lymphoplasmacytic lymphoma/Waldenstrom's macroglobulinemia (LPL/WM) as the underlying cause (7). Hemolysis is usually mild in CAD and has never been reported to cause renal hemosiderosis (8).

However, we encountered a patient with CAD and LPL/WM who developed acute kidney injury (AKI) caused by renal hemosiderosis due to severe intravascular hemolysis.

Case Report

A 67-year-old man was admitted to our hospital with malaise and jaundice in December 2013. He was a heavy drinker and had been drinking much more than usual for one week prior to admission. For the past three years, he had noted Raynaud's phenomenon and dark urine after exposure to cold. The results of his previous annual medical checkups were unremarkable, except for an elevation of total bilirubin to 2.0-3.5 mg/dL.

On admission, his blood pressure was 132/73 mmHg, pulse rate was 100/min, and body temperature was 39.5°C. A physical examination showed conjunctival pallor and severe jaundice, but there was no abdominal tenderness, splenomegaly, or lymphadenopathy. Laboratory tests revealed liver dysfunction, renal dysfunction, and hemolytic anemia (Table 1): GOT, 302 U/L; GPT, 266 U/L; gamma-glutamyl transpeptidase (γ-GTP), 440 U/L; alkaline phos-

[1]Nephrology Center, Toranomon Hospital, Japan, [2]Hematology Center, Toranomon Hospital, Japan, [3]Department of Pathology, Toranomon Hospital, Japan, [4]Department of Pathology, Yokohama City University, Graduate School of Medicine, Japan and [5]Okinaka Memorial Institute for Medical Research, Japan
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Correspondence to Dr. Yoshifumi Ubara, ubara@toranomon.gr.jp

Table 1. Laboratory Tests Revealed Liver Dysfunction, Renal Dysfunction, and Hemolytic Anemia.

		Normal range			Normal range
Blood tests					
WBC	9,300 /μL	3,400-9,200	CRP	6.4 mg/dL	0.0-0.3
Seg	84.5 %	45.6-73.2	Haptoglobin	<10 mg/dL	
Eos	0.0 %	0.6-8.4	Direct Coombs test	+	-
Lym	12.5 %	19.0-45.4	Anti-complement antibody	+	-
RBC	271×10^4 /μL	$400\text{-}566\times10^4$	Cold agglutinins	65,536	-
Hb	9.2 g/dL	13.0-17.0	IgG	1,395 mg/dL	870-1,700
Plt	25.1×10^4 /μL	14.1-32.7	IgA	298.1 mg/dL	110-410
TP	7.4 g/dL	6.9-8.4	IgM	282.6 mg/dL	35-220
Alb	4.3 g/dL	3.9-5.2	IgM-κ M protein	+	-
GOT	302 U/L	13-33	CH50	25 U/mL	30-50
GPT	266 U/L	8-42	C3	80 mg/dL	86-160
LDH	1,148 U/L	119-229	C4	7 mg/dL	17-45
ALP	310 U/L	117-350	Antinuclear antibody	<40	<40
γ-GTP	440 U/L	9-109	Rheumatoid factor	6 IU/mL	0-15
T-Bil	45.1 mg/dL	0.3-1.1	Anti-M2 antibody	<1.5 EU	<1.5
D-Bil	30.1 mg/dL	0.0-0.2	MPO-ANCA	<10 EU	<1.0
UN	24 mg/dL	8-12	PR3-ANCA	<10 EU	<1.0
Cr	1.39 mg/dL	0.65-1.06	Cryoglobulin	-	-
eGFR	40.5 mL/min		Soluble IL2 receptor	345 U/mL	145-519
Na	138 mEq/L	139-146	Total cholesterol	146 mg/dL	122-240
K	4.5 mEq/L	3.7-4.8	Triglyceride	77 mg/dL	30-150
Fe	198 μg/dL	80-120	Cholinesterase	186 IU/L	220-495
TIBC	263 μg/dL	253-383	Prothrombin time	62.5 %	>75
Ferritin	2,172 μg/L	10-190	APTT	22.5 s	25-36
Urine tests					
Protein	3.0 g/gCr	<0.15	Bilirubin	+	-
RBC	1-4 HPF	<1	Urobilinogen	2+	1+
			BJP-κ	+	-

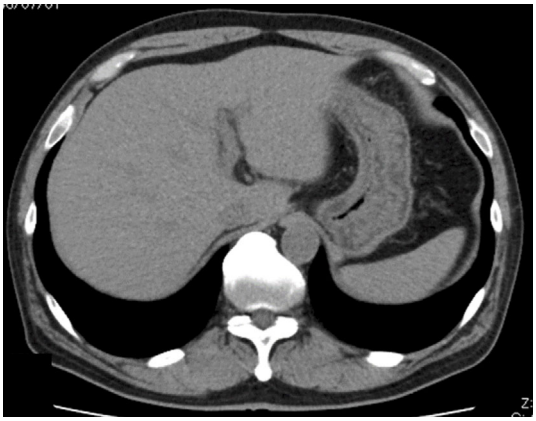

Figure 1. Non-contrast computed tomography showed no significant changes in the liver or biliary tract.

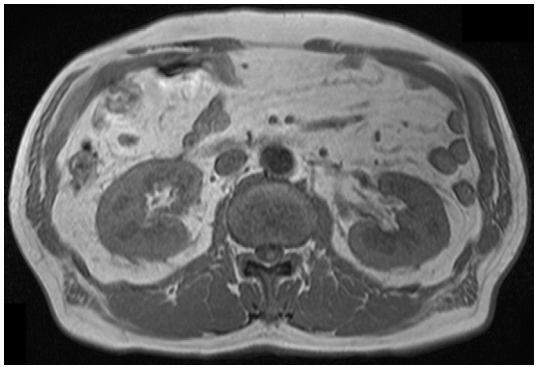

Figure 2. T2-weighted magnetic resonance imaging showed no specific signal intensity change in the kidneys.

phatase (ALP), 310 U/L; lactate dehydrogenase (LDH), 1148 U/L; total bilirubin (T-Bil), 45.1 mg/dL; direct bilirubin (D-Bil), 30.1 mg/dL; serum creatinine, 1.39 mg/dL; blood urea nitrogen, 24 mg/dL; urine protein, 3.0 g/gCre; urine blood (2+) with 1-4 red blood cells per high-power field; urine bilirubin (3+); and urine urobilinogen (2+). His hemoglobin was 9.2 g/dL, and haptoglobin was <10 mg/dL. The direct Coombs test and anti-complement antibody were both positive, and the cold agglutinin titer was 65,536. No evidence of infection was identified by blood cultures, viral serology (for hepatitis A virus, hepatitis B virus, hepatitis C virus, hepatitis E virus, Epstein-Barr virus, cytomegalovirus, and human immunodeficiency virus), or testing for *Mycoplasma*. Accordingly, primary CAD was diagnosed. Non-contrast computed tomography showed no significant changes in the liver or biliary tract. The kidney size was 11 cm, with no hydronephrosis in either kidney (Fig. 1). Mag-

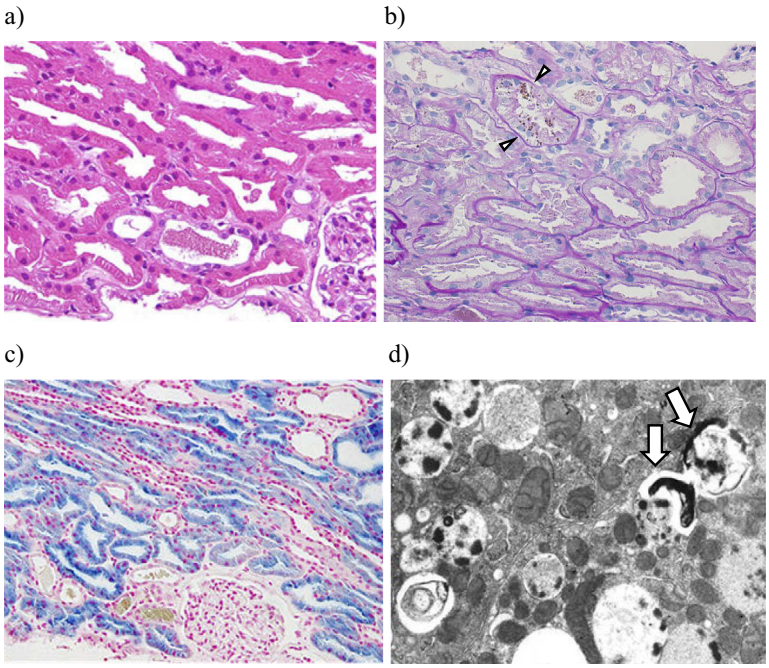

Figure 3. **Kidney pathology. (a) Hematoxylin and Eosin staining shows acute tubular necrosis. (b) Periodic acid-Schiff staining shows brown pigment in the proximal tubular epithelial cells (arrowhead). (c) Prussian blue staining shows numerous hemosiderin deposits in the proximal tubular epithelial cells. (d) Electron microscopy shows granular hemosiderin deposits in the lysosomes of proximal tubular epithelial cells.**

netic resonance cholangiopancreatography also showed no evidence of biliary obstruction and no specific signal changes in the kidneys (Fig. 2).

After admission, his liver function improved spontaneously, but anemia progressed, and the renal function also deteriorated, with the serum creatinine rising to 6.3 mg/dL on hospital day 2. At this time, we detected positive serum IgM-κ protein and urine Bence-Jones protein (BJP)-κ. Therefore, we performed a bone marrow biopsy and kidney biopsy on day 3.

Bone marrow biopsy findings

The bone marrow smear contained 1.4% large lymphoid cells and 0.6% plasma cells. Flow cytometry showed that the lymphoid cells were positive for CD5dim, CD19, CD20, CD22, CD23, and BCL2 and negative for CD10 and CyclinD1 along with light chain restriction (κ>λ), suggesting monoclonal proliferation of B cells. Erythrocytes did not express CD55 or CD59, excluding the possibility of PNH. These findings were compatible with a diagnosis of LPL/WM.

Kidney biopsy findings

The biopsy specimen contained 51 glomeruli. Only one glomerulus showed global sclerosis, and there were no significant changes in the other glomeruli. On hematoxylin and eosin staining, there was epithelial thinning, loss of the brush border, and detachment of epithelial cells in the proximal tubules, indicating acute tubular necrosis (Fig. 3a). In sections subjected to periodic acid-Schiff staining, deposits of brown pigment were seen in the proximal tubules (Fig. 3b), and Prussian blue staining revealed numerous hemosiderin deposits in the proximal tubular epithelial cells (Fig. 3c). Immunofluorescence was negative for immune deposits. Electron microscopy revealed granular hemosiderin deposits in the lysosomes of proximal tubular epithelial cells (Fig. 3d). Thus, the final diagnosis was AKI caused by renal hemosiderosis that was secondary to CAD due to underlying LPL/WM.

Clinical course

Fig. 4 summarizes the patient's clinical course. After the diagnosis of CAD, treatment with dexamethasone was started on hospital day 3 (40 mg/day for 4 days), and plasma exchange was initiated from day 4. We used fresh-frozen plasma as replacement solution, and the average volume of treated plasma was 3,400 mL for each session. However, his kidney function deteriorated rapidly on day 4 (with oliguria and serum creatinine of 7.2 mg/dL), requiring hemodialysis. Hemolysis improved rapidly after the initiation of dexamethasone therapy and plasma exchange, and kidney dysfunction also improved accordingly. Hemodialysis was withdrawn on hospital day 9 (after 3 sessions), and plasma exchange was withdrawn on day 11 (after 4 sessions). He started treatment with rituximab (375 mg/m^2 weekly) for LPL/WM on day 14 and received a total of 4 doses. The patient was discharged on hospital day 35 with a serum creatinine level of 1.0 mg/dL, hemoglobin of 7-8 g/dL, and total bilirubin of 2 mg/dL. Four years later, he is doing well without any treatment, but with a reduced alco-

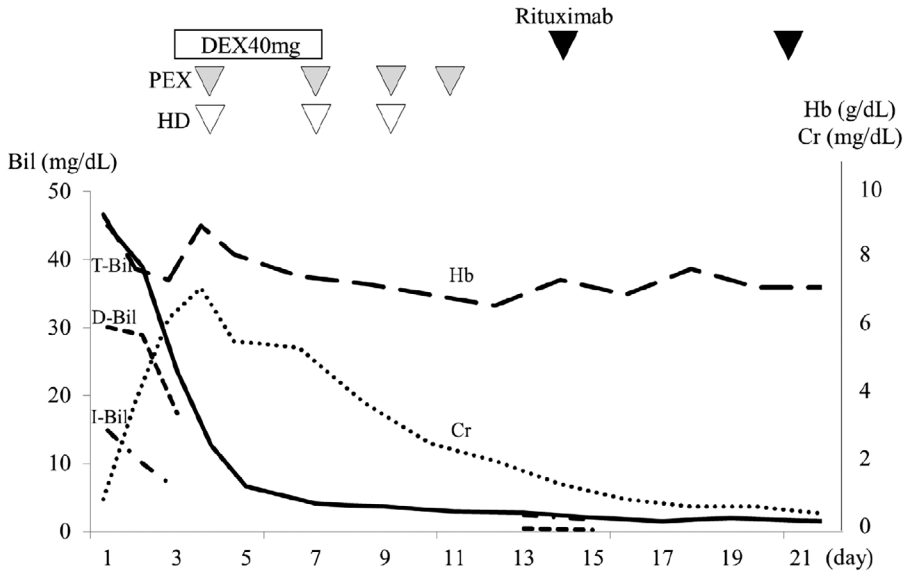

Figure 4. **Clinical course.**

Table 2. **The Patient's Current Blood Test Data 4 Years after Treatment.**

		Normal range			Normal range
Hb	11.6 g/dL	13.0-17.0	Haptoglobin	<10 mg/dL	
LDH	240 U/L	119-229	Direct Coombs test	+	-
T-Bil	3.0 mg/dL	0.3-1.1	Anti-complement antibody	+	-
D-Bil	0.0 mg/dL	0.0-0.2	Cold agglutinins	16,484	-
UN	16 mg/dL	8-12	IgM	322.3 mg/dL	35-220
Cr	0.76 mg/dL	0.65-1.06	IgM-κ M protein	+	-
eGFR	77.1 mL/min		CH50	9 U/mL	30-50
			C3	69 mg/dL	86-160
			C4	2 mg/dL	17-45

hol intake and avoidance of cold exposure (Table 2).

Discussion

This is the first report of AKI caused by renal hemosiderosis in a patient with CAD and LPL/WM.

PNH is well known as the main cause of renal hemosiderosis (1, 2), which leads to AKI and even chronic kidney disease in some patients (2, 9). However, few reports have described AKI secondary to renal hemosiderosis in patients with other hemolytic diseases. The mechanism underlying renal hemosiderosis has been reported as follows (10, 11): When hemolysis occurs, dimeric hemoglobin binds with haptoglobin in the plasma, after which haptoglobin-hemoglobin complexes are taken up and degraded by reticuloendothelial cells. However, plasma haptoglobin becomes saturated if there is massive hemolysis, allowing free dimeric hemoglobin to be filtered through the glomeruli and absorbed by the proximal tubules. In the tubular cells, hemoglobin dissociates into heme and globin, and heme proteins cause AKI through three mechanisms: 1) direct cytotoxicity, 2) decreased renal perfusion due to depletion of nitric oxide, and 3) cast nephropathy when casts are formed via the interaction of heme proteins with Tamm-Horsfall protein. Since we did not observe any cast formation, we believe that the kidney injury in the present case was caused mainly by direct cytotoxicity and decreased renal perfusion.

Magnetic resonance imaging is the only imaging modality to reveal hemosiderin deposition in the renal cortex in patients with PNH via the reversal of the normal cortical and medullary intensity on T1-weighted images (the cortex shows a lower signal intensity than the medulla) and with very low cortical intensity on T2-weighted images (12). In the present case, we did not note these typical changes on MRI findings. Renal hemosiderin deposition in the current case would have been milder than in patients with PNH who suffer from repeated episodes of severe hemolysis attack.

Among patients with autoimmune hemolytic anemia, 15% have primary CAD (6). A bone marrow biopsy reveals a clonal lymphoproliferative disorder in 75% of CAD patients, and half of these patients have LPL/WM (7). Conversely, 3% of patients with LPL/WM also have primary CAD (13). Raynaud's phenomenon is induced by cold temperatures in 90% of CAD patients, and 75% also show exacerbation of hemolysis with febrile illnesses (7). The median hemoglobin

is reported to be 9.2 g/dL (4.5-15.6), and the median total bilirubin level is 2.4 mg/dL (0.6-9.5) (7). Thus, primary CAD is associated with relatively mild hemolysis, and there have been no reports of renal hemosiderosis or AKI in this disease. In the present patient, primary CAD was associated with atypically severe intravascular hemolysis, which resulted in AKI due to renal hemosiderosis. Although why this patient developed severe hemolysis is unclear, his excessive alcohol intake and alcoholic liver dysfunction may have been involved. An excessive alcohol intake is known to trigger hemolysis in patients with PNH (14), and a chronic excessive alcohol consumption affects the structural integrity of red blood cell membranes due to increased oxidative stress (15, 16). Furthermore, hepatic synthesis of haptoglobin and processing of heme proteins might have been impaired in this patient due to his severe alcoholic liver dysfunction.

In conclusion, primary CAD can be associated with severe hemolysis, resulting in AKI due to renal hemosiderosis, especially in patients with an excessive alcohol intake.

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

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