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CASE REPORT

Two patients with severe alcoholic hepatitis accompanied by hypercytokinemia and granulocytic hyperelastasemia, successfully treated by intravenous infusion of urinastarine (Miraclid)

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Abstract Severe alcoholic hepatitis (SAH) is not simply a disease of the liver, but it also causes infection and multiple organ failure, and therefore carries an extremely poor prognosis. We report the successful treatment of two patients with SAH. Case 1: The patient was a 55-year-old man. He was a heavy drinker whose alcohol intake had increased for some 3 years to 1.8 L sake a day. Slight clouding of consciousness, fever, and jaundice were evident on his admission to our hospital. Laboratory data showed leukocytosis with a predominance of polymorphonuclear leukocytes, and a decline in coagulability. He tested negative for various hepatitis virus markers. With the diagnosis of SAH made, steroid pulse therapy and bilirubin adsorption therapy were administered. The jaundice improved and the interleukin-8 (IL-8) level decreased. Continuous intravenous infusion of urinastarine (Miraclid) normalized the granulocyte elastase level. Improvement was also seen in coagulability, ascites, icterus and consciousness. Case 2: The patient was a 49-year-old man. He was a heavy drinker whose alcohol intake had increased for 1 month. Fever, jaundice, ascites, and mild disturbance of consciousness were evident at the time of admission. Examination on admission diagnosed SAH. Bilirubin adsorption and continuous intravenous infusion of urinastarine were initiated. As a result, circulating IL-8 level was decreased and coagulability was improved. Therapy for granulocytic hyperelastasemia and hypercytokinemia supervening on SAH is a new effective approach on improvement in the disease.

Key words: cytokine, granulocyte elastase, severe alcoholic hepatitis, systemic inflammatory response syndrome, urinastarine.

INTRODUCTION

Severe alcoholic hepatitis (SAH) is not simply alcoholic liver disease; it also causes damage to other major organs, and carries an extremely poor prognosis.[1,2] Corticosteroids,[3–5] glucagon and insulin,[6] plasma exchange, leukoapheresis, and liver transplant[7] are currently used to treat SAH, but these methods have many drawbacks in terms of efficacy, convenience, and cost. Furthermore, the pathogenesis of the disease is not well understood. It can therefore be said that there is presently no effective established treatment for the disease. We report two patients with SAH who showed marked symptomatic improvement following treatment with urinastarine (urinary trypsin inhibitor (UTI); Miraclid, Mochida Pharm. Co. Ltd, Tokyo, Japan), focusing on neutrophilia and neutrophilic infiltration, findings specific to the disease, and hyperelastasemia and granulocytic hypercytokinemia supervening on SAH.

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CASE 1

The patient was a 55-year-old man whose chief complaints were jaundice and generalized fatiguability. The patient had had hyperuricemia since the age of 46 years and had a non-contributory family history. He also had no history of transfusions.

Present illness

The patient was a habitual drinker (previously consuming approximately 0.9 L of alcohol in sake equivalent per day). From approximately 1993, his alcohol consumption increased to approximately 1.8 L sake (one of the Japanese liquor categories) per day. He developed jaundice in March 1998, but did not seek treatment. He had his last drink on 5 May of the same year, after experiencing increasing generalized fatiguability. He presented at a local hospital (Suibaragho Hospital, Niigata, Japan) on 11 May 1998, after the jaundice was pointed out by family members. He was admitted to the same hospital for a suspicion of alcoholic hepatitis, and treated conservatively. However, the jaundice failed to improve and coagulability decreased. He was subsequently referred to the Department of Gastroenterology of the Saiseikai Niigata Second Hospital and admitted on 26 May 1998.

Status on admission

The patient was 160 cm tall and weighed 75 kg. The pulse was 75 and regular, temperature was 37.7°C, blood pressure was 108/64 mmHg, and the respiration rate was 26. Slight clouding of consciousness (Sherlock Coma Scale grade I) was evident. An examination of the ocular conjunctiva showed jaundice. Swelling caused by marked deposition of subcutaneous fat was observed on the neck, and from the nape of the neck to the back. Thoracic findings were normal. The liver edge was palpable four finger-breadths between the right costal arches above the mid-clavicular line, but no tenderness was present. Mild ascites and edema in the lower extremities were also evident.

Laboratory findings on admission

Total bilirubin was 23.0 mg/dL, indicating marked jaundice. The transaminase levels were elevated, with a predominance of AST, and leukocytosis (13 700/mm^3) with a predominance of polymorphonuclear leukocytes (88%) was noted. The prothrombin time (PT) was 35.5% and the Hepaplastin Test (HPT) value was 27.7%, indicating a marked coagulation disorder. Blood urea nitrogen (BUN) was 68 mg/dL and creatinine (Cr) 1.5 mg/dL, both signs of renal failure. Tests for various hepatitis virus markers for hepatitis A virus, hepatitis B virus and hepatitis C virus were negative, as well as antinuclear antibodies and antimitochondrial antibodies.

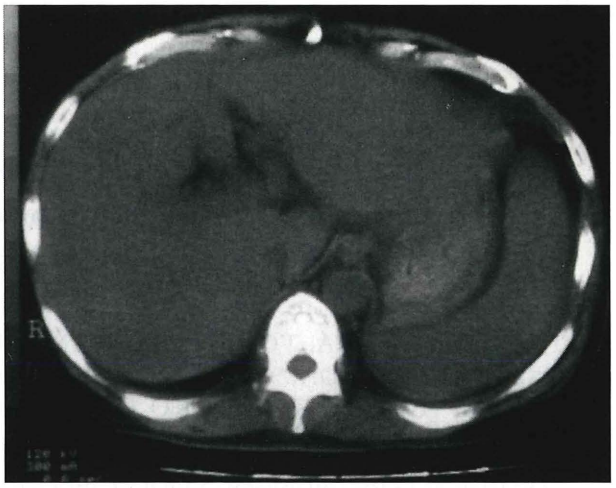

Figure 1 Abdominal CT. An abdominal CT scan revealed marked hepatosplenomegaly and mild ascites.

Abdominal CT (Fig. 1) showed marked hepatosplenomegaly and mild ascites. An ultrasound-guided liver biopsy (Fig. 2) revealed fibrosis, but this had not progressed to cirrhosis. Advanced hepatocyte loss and necrosis were also evident. Bile thrombi and hyaline bodies were present, and marked infiltration of polymorphonuclear leukocytes was observed. The patient was diagnosed as having SAH based on these findings, and treatment was started.

Progress after admission

Steroid pulse therapy and bilirubin adsorption were initiated because cessation of alcohol consumption, fluid replacement, and administration of liver-supporting agents were without effect at the previous hospital (Fig. 3) After three courses of bilirubin adsorption therapy, the total bilirubin concentration decreased to 8.8 mg/dL, but polymorphonuclear leukocytosis persisted. There was no increase in concentrations of α-fetoprotein (AFP) (2.2 mg/mL) or hepatocyte growth factor (HGF) (0.95 ng/mL), nor was there a rise in the endotoxin level (10 pg/mL) before methicillin-resistant *Staphylococcus aureus* (MRSA) septicemia. No increase was observed in concentrations of the tumor necrosis factor-α (TNF-α) and interleukin-6 (IL-6) (18 mg/L) as inflammatory cytokines, but IL-8 was elevated to 47.1 pg/mL. Granulocyte elastase (polymorphonuclear leukocyte elastase, or PMNE) was increased to 510 μg/L. Although bacterial infection was absent, there was concern about the possibility of progression to multiple organ failure associated with the augmentation of the systemic inflammatory response, particularly a rise in the granulocyte elastase level. Elevation of granulocyte elastase was treated by continuous intravenous infusion of 300 000 units of urinastarine for 5 days. As a result of treatment, the granulocyte elastase level normalized to 65 mg/L, coagulation improved dramatically, as indicated by an increase in PT to 84.5% and HPT

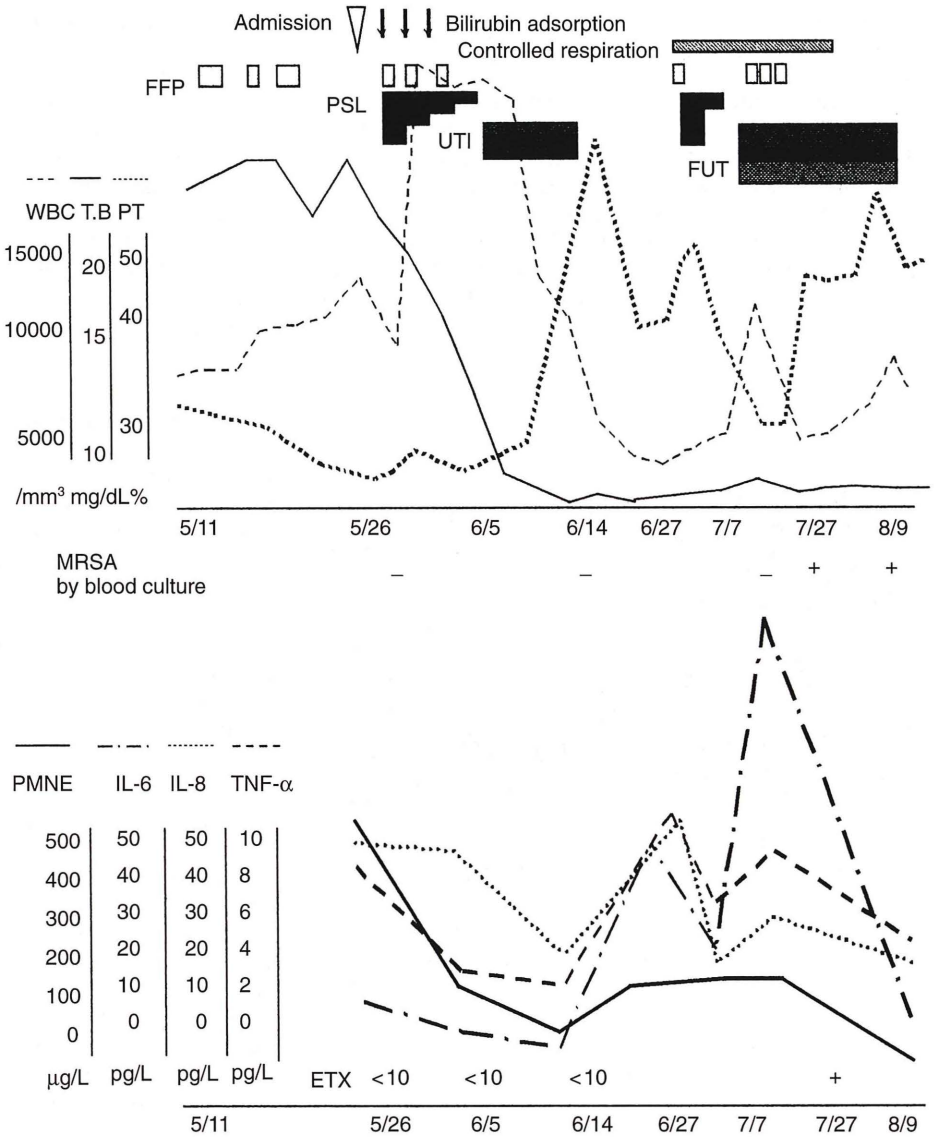

Figure 2 Progress after admission (case 1). WBC, white blood cells; TB, total bilirubin; PT, prothrombin time; MRSA, methicillin-resistant *Staphylococcus aureus*; FFP, fresh frozen plasma; PSL, predonisolone; UTI, urinary trypsin inhibitor; FUT, nafamostat mesilate; PMNE, polymorphonuclear leukocyte elastase; IL, interleukin; TNF, tumore necrosis factor; ETX, endotoxin. Shaded boxes, intravenous infusion of UTI; blank boxes, FFP; waved boxes, intravenous infusion of PSL; arrows, bilirubin absorption; hatched boxes, period of the controlled respiration; dotted boxes, intravenous infusion of FUT.

to 67.9%, and general physical status improved temporarily.

However, after completion of urinastarine, the white blood cell (WBC) count rose to 12 500 (neutrophils 94%), coagulability function declined, and the PMNE level increased once more. Respiratory status worsened, and chest radiographs showed frosted glass shadows extending over all pulmonary fields. Oxygen tension (PO_2) was 44.5 mmHg, carbon dioxide tension (PCO_2) 17.2 mmHg, O_2 saturation (O_2SAT) 83.8, pH 7.416, HCO^{3-} 11.0 mEq/L, and base excess (BE) −11.3 mmol/L, indicating progression to acute respiratory distress syndrome (ARDS). The patient was intubated and placed on controlled respiration. Steroid pulse therapy was resumed. As a result, there was improvement in the inflammatory findings and respiratory status, as indicated by a PO_2 of 87.1 mmHg, PCO_2 of 13.8 mmHg, O_2SAT of 96.5, pH of 7.34, HCO^{3-} of 7.6 mEq/L, and BE of −15.5 mmo/L. The patient resumed eating, and after persistent MRSA septicemia, confirming improvement in hepatic function and stabilization of general physical status, rehabilitation was started for muscle weakness associated with long-term confinement to bed. The patient was then discharged.

CASE 2

A 49-year-old man's chief complaint was jaundice and generalized fatiguability. He had a non-contributory family history and had no history of transfusions.

Present illness

The patient was a habitual drinker (previously consuming approximately 0.9 L alcohol in sake equivalent per day). From approximately June 1998, he drank all day long, consuming approximately 1–2 L beer, in addi-

tion to his usual sake consumption. The patient had his last drink on 29 June, 1998 after experiencing worsening generalized fatigue.

He was examined at a neighborhood hospital (Murakami General Hospital, Niigata, Japan) on 3 July, 1998, and was admitted there with a clinical suspicion of alcoholic hepatitis. Despite conservative treatment, the jaundice failed to improve, and the fever worsened with a decrease in coagulability. He was subsequently referred to the Department of Gastroenterology of the Saiseikai Niigata Second Hospital and admitted on 13 July, 1998.

Status on admission

The patient was 160 cm tall and weighed 72 kg. His pulse was 92 and regular, temperature 38.6°C, blood pressure 118/70 mmHg, and he had a respiration rate of 27/min. Consciousness was clear. Examination of the ocular conjunctiva showed jaundice. Thoracic findings were normal. The liver edge was palpable three finger-breadths between the right costal arches above the mid-clavicular line, but no tenderness was present. Ascites and edema in the lower extremities were evident.

Laboratory findings on admission

Levels of transaminases, particularly AST, were elevated and total bilirubin was 13.3 mg/dL, indicating severe jaundice. Leukocytosis (13 800/mm^3) with a predominance of polymorphonuclear leukocytes (87%) was evident. Prothrombin time was 30.1% and HPT 20.7%, indicating a marked decrease in coagulability. α-Fetoprotein was increased to 15.5 ng/mL. The patient tested negative for various hepatitis virus markers, and antinuclear antibodies and antimitochondrial antibodies.

Progress after admission

The patient was diagnosed as having SAH based on the negative tests for various hepatitis virus markers and autoantibodies, the absence of prior oral drug use, and a history of heavy alcohol consumption (Fig. 4). Bilirubin adsorption therapy and continuous intravenous infusion of 300 000 units of urinastarine were performed for 24 days, as cessation of alcohol consumption, fluid replacement, and administration of liver-supporting agents initiated were without effect at

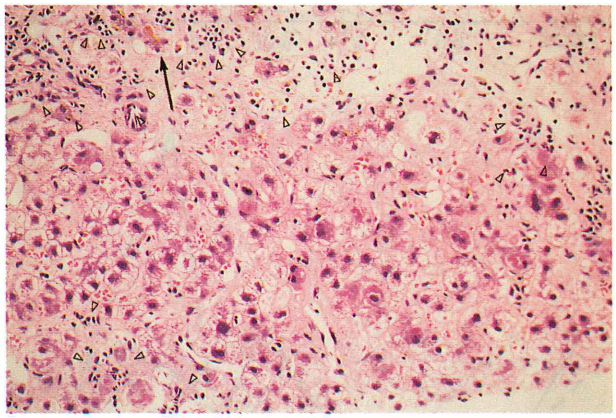

Figure 3 Liver biopsy tissue. An ultrasound-guided liver biopsy showed fibrosis, but this had not progressed to cirrhosis. Advanced hepatocyte loss and necrosis were evident. Bile thrombi and hyaline bodies were present, and marked polymorphonuclear leukocyte infiltration was observed. Cholestasis is shown by an arrow and neutrophil infiltration is shown by arrowheads.

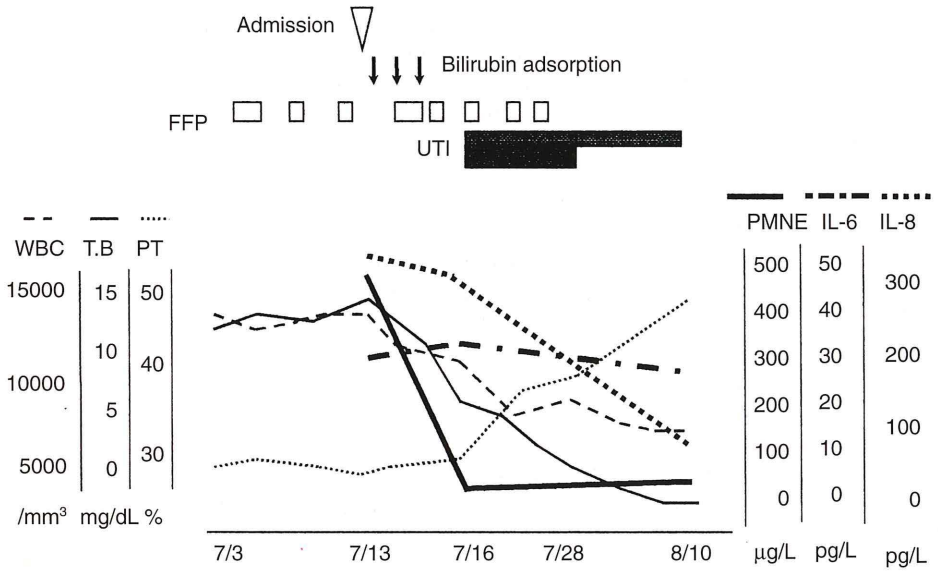

Figure 4 Progress after admission (case 2). WBC, white blood cells; TB, total bilirubin; PT, prothrombin time; PMNE, polymorphonuclear leukocyte elastase; IL, interleukin; FFP, fresh frozen plasma; UTI, urinary trypsin inhibitor. Shaded areas, intravenous infusion of UTI; blank boxes, FFP; arrows, bilirubin absorption.

the previous hospital. After five courses of bilirubin adsorption therapy, the total bilirubin concentration decreased to 6.8 mg/dL, and the PMNE level normalized. After intravenous infusion of urinastarine, the polymorphonuclear leukocytosis improved, inflammatory cytokine levels decreased, and PT improved dramatically. The patient's general physical status also improved. Thereupon, he was discharged.

DISCUSSION

Alcoholic hepatitis refers to a disease entity characterized by clinical symptoms of acute liver injury. Histology of this disease is hepatocellular necrosis and ballooning, the presence of Mallory bodies and polymorphonuclear cell infiltration in the liver. Heavy drinkers are precipitated into SAH by increased excessive alcohol consumption. The disease varies in severity from mild forms that rapidly improve following abstinence from alcohol and introduction of an appropriate diet, to severe forms similar to acute hepatic failure and multiple organ failure, which can result in death 1–2 months after onset.

It was suspected early that the pathogenesis of severe alcoholic hepatitis with a poor prognosis differed from that of ordinary alcoholic hepatitis. Hawkins[8] referred to this disease state as 'fulminant alcoholic hepatitis', while Theodossi *et al.* [3] and Halle *et al.* [2] used the term 'severe acute alcoholic hepatitis' in an attempt to classify the more severe forms. Phillips and Davidson[1] omitted the word 'acute' and instead proposed 'severe alcoholic hepatitis' (SAH), because fibrosis of varying degrees is always seen in patients with the disease caused by chronic hepatic lesions and long-term immoderate alcohol ingestion. The last term has come into widespread use.

Severe alcoholic hepatitis is characterized by a marked polymorphonuclear leukocytosis, which is also referred to as a diagnostic criterion, and marked elevation in the WBC count when the patient is first hospitalized; no infection has yet developed. Wallach and Jacob suggested the involvement of a humoral factor (colony stimulating factor, CHF) that promotes production of granulocytes, as the WBC count tends to be elevated in patients who die, and therefore appears to reflect prognosis.[9] The cause of the leukocytosis in SAH is unclear. It appears that TNF-α and IL-8 induced by endotoxin cause neutrophilia in alcoholic hepatitis, and that activation of neutrophils by IL-8 also plays a role.[10–13]

Inflammation is currently evaluated by measuring circulating IL-6 and IL-8 levels because the sensitivity and accuracy of measuring circulating IL-6 and IL-8 levels are improved. This is based on the fact that IL-1 and TNF are induced locally, and measuring levels of cytokines that induce the primary biological defense response provides an indication of the severity of local damage. We investigated treatment of SAH by measuring PMNE, cytokine, and endotoxin levels, and removing and adjusting levels of these substances which are considered to contribute to hepatic necrosis.[14]

In our patients, the diagnostic criteria for systemic inflammatory response syndrome (SIRS) were compatible.[15] Severe alcoholic hepatitis does not simply involve liver failure; SIRS is also present. Once neutrophils have accumulated, infection and transportal translocation of bacteria and endotoxin occur. This leads to a second attack by reinducing cytokines that act on neutrophils that have accumulated in the major organs.[16] Treatment of this secondary multiple organ failure and ARDS requires accurate evaluation and treatment of SIRS. In case 1, it is suggested that neutrophil activation occurred as a result of priming and triggering after completion of urinastarine administration, leading to ARDS. In the SIRS, in which pro-inflammatory cytokines predominate, anti-inflammatory cytokines are induced simultaneously, resulting in the compensatory anti-inflammatory response syndrome (CARS) characterized by a predominance of anti-inflammatory cytokines.[17] Susceptibility to infection is dependent on the balance between the pro-inflammatory and anti-inflammatory responses. We have no measurement of anti-inflammatory factors such as IL-10. In case 1, CARS was suspected to have played a role in the persistent MRSA septicemia that appeared after treatment for ARDS. Further study must look more carefully at the anti-inflammatory factors. It is reported that the onset of organ failure caused by the second attack after neutrophilic infiltration can be reduced by simply inhibiting PMNE activity.[18] It is therefore possible that administration of large quantities of urinastarine after neutrophilic infiltration inhibited the second attack in case 2. Inhibition of PMNE is of particular importance in the treatment of SAH, as it reduces production of IL-8.[19]

Amino acid sequence analysis shows that UTI is a proteoglycan with a molecular weight of 30 000 Daltons, which has two similar domains, and is linked to two carbohydrate chains.[20] Domain I appears to inhibit chymotrypsin and PMNE, and domain II to inhibit chymotrypsin, plasmin, and trypsin. In addition, we speculate that UTI decreased cytokine-induced neutrophil chemoattractant after the first attack of SIRS.[21] Inter-α-trypsin inhibitor (ITI) is a precursor of UTI produced in the liver. It is degraded locally by PMNE, producing UTI which, in turn, reacts with protease.[22] If sufficient levels of inhibitors of proteases such as PMNE are present, organ failure does not occur. However, patients with reduced hepatic ITI production, as seen in SAH, are susceptible to organ failure caused by a shortage of protease inhibitors. Treatment of SAH therefore requires supplementation with UTI.

The future treatment of SAH requires a new approach that views the disease as a syndrome (SIRS) and takes into consideration the balance between the pro-inflammatory and anti-inflammatory responses at the onset of multiple organ failure, rather than focusing only on hepatic failure.

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