Case Report:
Unusual Warm Autoimmune Hemolytic Anemia in Non-Alcoholic Steatohepatitis

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Abstract. Warm autoimmune hemolytic anemia (WAIHA), a rare disease (0.2-1 per 100,000 population), ranges from an indolent form with mild hemolysis to a life-threatening condition that necessitates transfusion of incompatible red cells. WAIHA can be either idiopathic or secondary to medications or to a lymphoproliferative disorder. We report a case of profound hemolytic anemia in a liver-transplant eligible patient who was diagnosed with cirrhosis secondary to non-alcoholic steatohepatitis (NASH). The patient initially was treated with red cell transfusion, iv immunoglobulin, and steroids. He developed acute renal failure that required dialysis. Subsequent management included plasmapheresis and rituximab therapy. The patient developed hepato-renal syndrome and died from progressive hepatic failure. To our knowledge, this is the first report of an association between NASH and WAIHA.

Keywords: non-alcoholic steatohepatitis, autoimmune hemolytic anemia, hepatic cirrhosis

Introduction

Warm autoimmune hemolytic anemia (WAIHA) results from the production of autoantibody against unspecified high frequency red cell antigen(s) [1]. Usually, the antibody is of the IgG isotype and causes the destruction of patient and donor red cells at body temperature. Primary and secondary forms of the disease exist [1,2]. The activation of the immune system in primary cases is poorly understood. In WAIHA, the antigens targeted by the autoantibodies are either unclassified or react against red cell membrane protein(s) [3,4].

Non-alcoholic steatohepatitis (NASH) is the most common form of chronic liver disease and appears to be increasing in incidence [5,6]. The pathogenesis of NASH is poorly understood but evidently is multifactorial [7]. Possible pathogenic factors include carbohydrate- and insulin-excess, leading to TNF-mediated injury and formation of oxygen radicals, which can cause lipid peroxidation and mitochondrial dysfunction [6]. A small percentage of patients with NASH develop liver cirrhosis [8,9]. In one study, 23% of patients with NASH had concurrent extrahepatic immune diseases including diabetic mellitus, thyroiditis, immune (idiopathic) thrombocytopenia purpura, pericarditis, Sjogren’s syndrome, and synovitis [10,11]. Here we report a patient with a combination of WAIHA and NASH to alert physicians to a possible association between these two diseases.

Case Report

A 60-yr-old Caucasian non-obese man with a history of diabetes mellitus and no history of alcoholism presented with a 3-yr history of abnormal liver function tests and hepatic cirrhosis. Liver biopsy showed cirrhosis with portal and perisephal chronic inflammation with few eosinophils,
spotty acinar hepatocellular necrosis with ballooned hepatocytes, apoptotic bodies, and focal steatosis [Fig. 1]. An iron stain revealed mild reticuloendothelial and hepatocellular hemosiderosis, but HFE gene analysis for hereditary familial hemochromatosis was negative. A periodic acid-Schiff’s stain with diastase was negative, excluding alpha-1 anti-trypsin deficiency. No definitive Mallory bodies were seen by H&E stain, but Masson trichrome stain showed bridging and pericellular fibrosis (Fig. 1). None of the patient’s medications have known associations with this histological pattern of liver injury.

Serologic studies were negative for HCV antibody, HBsAg, HBsAb, HBeAg, HBeAb, HBC (hepatitis B core), total and HBC-IgM and HAV IgM, hepatitis delta antibody (HDVAB), HIV antibody, mycoplasma antibody IgM, CMV IgM antibody, or Treponema pallidum antibody. Serologic tests for anti-nuclear antibodies (ANA), anti-microsomal antibody (AMA), anti-ribonucleoprotein (RNP) antibody, anti-Smith antigen (Sm) antibody, antiparietal cell antibody, and anti-liver/kidney microsomal antibody (LKM) were all negative. The hepatic biopsy findings in combination with the serologic studies were most consistent with non-alcoholic steatohepatitis.

The patient came to our hospital with a 2-wk history of progressive fatigue and syncope. Laboratory test results on admission were: blood hemoglobin 4.8 gm/dl, Hct 13.7%, MCV 136.7 fl, reticulocytes 32%, platelets 92x10^3/µl, WBC 7.25

Fig. 1. Panel A: patient’s serum shows hemolysis on the admission day; Panel B: blood smear (modified Wright’s stain) shows anemia, anisocytosis, polychromasia, and spherocytosis (x1000); Panel C: blood smear stained with brilliant cresyl blue (reticulum stain) shows residual RNA/DNA in immature erythrocytes (x1000); Panel D: liver biopsy (Masson’s-trichrome stain) shows bands of fibrous tissue surrounding hepatocytes with macro- and micro-steatosis (x100).
x10^3/µl, serum glucose 277 mg/dl, alanine aminotransferase (ALT) 31 IU/L, aspartate aminotransferase (AST) 130 IU/L, haptoglobin <20 mg/dl (normal 90-250), urea nitrogen 37 mg/dl, creatinine 1.7 mg/dl, glomerular filtration rate (GFR) >60 ml/min, lactate dehydrogenate (LD) 1512 IU/L (normal 90-250), serum bilirubin 10.9 mg/dl (normal 0.3-1.3), and serum direct bilirubin 4 mg/dl (normal 0.0-0.5).

There was no recent history of blood transfusion. The patient’s plasma was dark red, indicating hemolysis (Fig. 1). Blood smears showed anemia, polychromasia, and spherocytosis (Fig. 1). Red cell antibody screening was strongly positive (3+) at the immunoglobulin phase and weakly positive (1+) at the immediate spin. Direct antiglobulin test was strongly positive (3+) for IgG and negative for complement C3. Indirect antiglobulin test was positive for the entire 16-red cell panel. When the patient’s red cells were treated with commercial acid solution to dissociate the binding autoantibodies, the eluate was positive (3+) with all red cells tested. There were no detectable anti-red cell alloantibodies.

The patient’s therapy initially consisted of Solu-Medrol (1 mg/kg/day) and standard doses of iv immunoglobulins (IVIG) for 3 consecutive days. On the third day of IVIG administration, the serum creatinine level peaked at 3.9 mg/dl. Subsequently, the patient underwent hemodialysis every other day for 6 days. The serum LD activity decreased to 700 IU/L, but the patient remained transfusion-dependent. The patient received daily plasmapheresis for 3 days using plasma and 5% serum albumin (50/50, v/v) as replacement fluid, followed by a single dose of rituximab (375 mg/m^2). The patient required multiple red cell transfusions and received 8 units of incompatible packed red cells during his hospital stay, with an increase of blood Hb level to 8 gm/dl. His clinical condition improved and he was discharged from the hospital. Two days later, he was readmitted with confusion, worsening jaundice, and abdominal bloating. His liver failure progressed rapidly as the serum bilirubin increased to 70 mg/dl. The autoimmune hemolytic anemia also worsened despite transfusion support and steroid therapy. The patient died two days after re-admission.

**Discussion**

Warm autoimmune hemolytic anemia (WAIHA) results from production of an IgG isotype autoantibody against unclassified red cell antigens or red cell membrane proteins [3,4]. In most cases of WAIHA, the nature of the antigens is unknown [12]. WAIHA is often treated with a variety of immunosuppressive agents [13,14]. Recently rituximab has been successful in the treatment of WAIHA [15]. Warm autoimmune hemolytic anemia has been reported in patients with several liver diseases including chronic hepatitis C virus infection [16,17], hepatitis A viral infection [18], and asymptomatic carrier of hepatitis B viral infection [19], primary biliary cirrhosis [20], and liver transplant recipients for primary biliary cirrhosis [21]. Autoantibodies including anti-nuclear and anti-smooth muscle antibodies have been detected in some liver diseases [10,11]. However, these autoantibodies are not associated with severe degrees of injury on liver biopsy. The lack of concordance between the presence of autoantibodies and the severity of injury on liver biopsy does not preclude a relationship between the liver diseases and development of autoimmune diseases. Non-alcoholic fatty liver disease (NAFLD) and non-alcoholic steatohepatitis (NASH) are two terms used to describe a similar disease. NASH is the most common cause of incidental serum elevations of liver enzyme activities in North America and Europe [22]. Although originally considered a benign entity, the spectrum of NASH is variable, ranging from steatosis, with a benign prognosis, to steatohepatitis and cirrhosis [22].

Our patient was diagnosed as having NASH with advanced cirrhosis and with negative serologic tests for ANA and AMA, including antibodies to liver/kidney microsomes type 1 (anti-LKM), excluding autoimmune hepatitis [23]. While the patient was waiting for a liver transplant he developed autoimmune hemolytic anemia. He did not respond to immune suppression treatment, to immuno-modifiers such as IVIG, or to plasma apheresis and he died secondary to liver failure. No autopsy was done. To our knowledge this is the first reported case of NASH in combination with warm auto-immune hemolytic anemia.
References


