A Case of Cryoglobulinemia Associated with Nonalcoholic Steatohepatitis

David Giangreco, MD[1]
Steven J. Scaglione, MD[2]
Stanley Martin Cohen, MD[2]
Rodney Tehrani, MD[3]

[1]Department of Internal Medicine, [2]Department of Gastroenterology and Hepatology, [3]Department of Rheumatology, Loyola University Medical Center, Maywood, Illinois

Case Report

A 44-year-old male with a history of hypertension and abnormal liver function test results presented to his primary care physician with a 6-week history of bilateral lower extremity rash, arthralgias, subjective fevers, and chills. His symptoms began with a diffuse, erythematous rash that involved his trunk, arms, and legs but spared his palms, soles, and face. He was referred to a dermatologist and was diagnosed with contact dermatitis. He was treated with an 80-mg injection of methylprednisolone acetate (Depomedrol, Pharmacia and Upjohn) and achieved complete resolution of the rash within 3 days. Six weeks later, the rash reappeared, again accompanied by diffuse myalgias, arthralgias in the patient's hands and elbows, and flu-like symptoms, including fever and chills. He was treated with moxifloxacin for presumed community-acquired pneumonia. When his symptoms did not resolve after 1 week of antibiotic therapy, he was referred to a rheumatologist, and further work-up was begun.

The patient's medical history was significant for hypertension, mildly elevated liver function test results, seasonal allergies, and recent pneumonia. His only medications were fluticasone propionate (Flonase, GlaxoSmithKline) and moxifloxacin. He denied tobacco, alcohol, or illicit drug use. He had no family history of liver disease or autoimmune disease. His physical examination on presentation was significant for a diffuse, bilateral rash with erythematous macules and purpuric papules on his lower extremities.

Address correspondence to:
Dr. Steven J. Scaglione, Department of Gastroenterology and Hepatology, Loyola University Medical Center, 2160 South First Avenue, Fahey Center, Building 54, Maywood, IL 60153; Tel: 708-216-0464; Fax: 708-216-4113; E-mail: stevescaglione@gmail.com

Laboratory evaluation was significant for an aspartate aminotransferase level of 96 IU/L and an alanine aminotransferase level of 112 IU/L. Serologic tests for hepatitis B virus, hepatitis C virus (HCV), ceruloplasmin, α-1 antitrypsin, anti–liver-kidney microsome-1, and hemochromatosis were negative. Testing for HCV RNA was negative. Tests for antinuclear antibody and anti–smooth muscle antibody were both low-titer positive at 1:40. Testing for rheumatoid factor was positive at a level of 51 IU/mL. Results of an extractable nuclear antigen panel—including ribonucleoprotein-smith, anti–Sjögren syndrome A, anti–Sjögren syndrome B, SCL-70, and anti–Jo-1—were negative, as were peripheral antineutrophil cytoplasmic antibody testing results and anticardiolipins. The patient's complement 4 level was 2 mg/dL (normal, 16–38 mg/dL), and his complement 3 level was 75 mg/dL (normal, 75–152 mg/dL). Immunoglobulins, light chains, and serum protein electrophoresis were normal. Cryoglobulins were present in the serum consistent with type III cryoglobulinemia. A biopsy of the left thigh revealed superficial acute leukocytoclastic vasculitis. A liver biopsy revealed mixed microvesicular and macrovesicular steatosis involving 80% of the hepatic parenchyma with bridging fibrosis, mild portal chronic inflammation, and balloon-cell changes consistent with nonalcoholic steatohepatitis (NASH; Figures 1 and 2).

The patient was diagnosed with type III cryoglobulinemia and was treated with prednisone at a dose of 40 mg daily. His symptoms improved rapidly over the following week. Corticosteroids were tapered within a 2-week period, and he was started on mycophenolate mofetil at a dose of 1 g twice daily. After having been treated with mycophenolate mofetil for 1 year without any recurrence of symptoms, he opted to stop taking this medication without consulting a physician. One year later, he

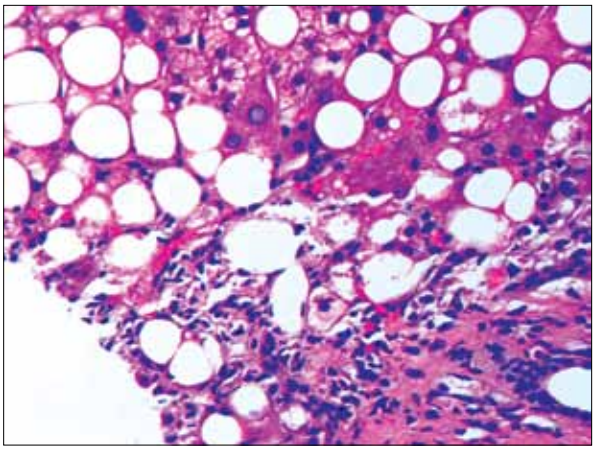

Figure 1. Hematoxylin and eosin stain of the liver biopsy (400× magnification).

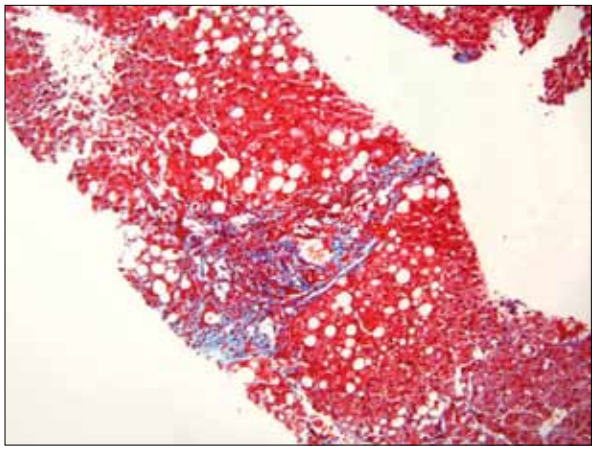

Figure 2. Trichrome stain of the liver biopsy (100× magnification).

developed recurrence of purpuric rash and arthralgias. Reinitiation of immunosuppression with a 2-week course of prednisone starting at a dose of 40 mg daily and tapering by 10 mg every 5 days resulted in resolution of these symptoms. At that time, he was restarted on mycophenolate mofetil at a lower dose (500 mg twice daily).

After 1 year without recurrence of symptoms, the decision was made to decrease the mycophenolate mofetil dose to 500 mg daily. He had no further episodes of disease recurrence after that time. The dose of mycophenolate mofetil was then slowly decreased over the following 2 years; the dose was reduced to 250 mg daily after 9 months, and mycophenolate mofetil was stopped completely 6 months thereafter. He has now remained without symptoms and off mycophenolate mofetil for almost 1 year.

In regard to his liver disease, the patient has remained at a weight of 220 lbs since the diagnosis of NASH was made. Three years after his initial confirmatory liver biopsy, he underwent an open cholecystectomy, and gross examination noted irregular contours and light coloring suggestive of fatty infiltration. A biopsy was obtained at that time and again revealed steatohepatitis with bridging fibrosis. Macrovesicular changes with associated balloon-cell formation were seen in 70–75% of the biopsy. At the time of this biopsy, the patient denied any alcohol use and continued to consume a high-fat, high-cholesterol diet. He also continued to have impaired fasting glucose. He was encouraged to remain completely abstinent from alcohol and was started on vitamin E at a dose of 400 IU twice daily. He was referred to the nutrition clinic and for endocrinology consultation at that time. He continues to be followed closely by the hepatology and rheumatology clinics.

Discussion

Cryoglobulins are immune-complex proteins that are soluble when heated and precipitate when cooled. There are 3 classes of cryoglobulins: type I (monoclonal), type II (mixed monoclonal and polyclonal), and type III (polyclonal). When cryoglobulins are not associated with a recognizable disease, they are designated as idiopathic or essential. Type I cryoglobulinemia is associated with macroglobulinemia, multiple myeloma, or monoclonal gammopathy of undetermined significance. Type II cryoglobulinemia is most commonly associated with HCV infection, in addition to lymphoproliferative diseases, glomerulonephritides, and chronic infections. Type III cryoglobulinemia is also associated with HCV infection. These immune complexes can precipitate and deposit on vascular endothelium. The most commonly affected organs are the skin, kidneys, and peripheral nerves. The most common clinical presentation involves palpable purpura, but cryoglobulinemia can also affect the small and medium-sized vessels of the kidney and peripheral nerves, leading to membranoproliferative glomerulonephritis or peripheral neuropathy.[1]

In patients with mixed cryoglobulinemia who are HCV-positive, the first-line treatment would be to attempt to achieve sustained virologic response with HCV treatment, usually pegylated interferon and ribavirin. Immunosuppression with rituximab (Rituxan, Genentech/Idec Pharmaceuticals), prednisone, or mycophenolate mofetil can be used in patients with persistent symptoms of cryoglobulinemia who do not achieve sustained virologic response.[2]

The prevalence of overweight and obesity has increased markedly since the mid-1970s. According to the Centers for Disease Control and Prevention and

data from the National Health and Nutrition Examination Survey, the prevalence of obesity has increased from 15% in 1976–1980 to 32% in 2003–2004.[3] Nonalcoholic fatty-liver disease (NAFLD) has been increasingly recognized among patients with features of metabolic syndrome, including central obesity, insulin resistance, hypertension, and hyperlipidemia.[4-7] NAFLD is the most common form of chronic liver disease in developed countries.[8] Given its association with rising rates of obesity, NAFLD has become an important public health concern.

NASH is associated with increases in both cardiovascular and liver-related mortality.[9,10] Estimates suggest that 71 million Americans have hepatic steatosis. Furthermore, 20% of all new office visits for chronic liver disease are related to NAFLD. NASH is characterized by the presence of steatohepatitis, lobular inflammation, and balloon-cell degeneration. Although it is unknown how many patients with NAFLD will progress to NASH, several studies have evaluated fibrosis progression via liver biopsy. In 2005, Adams and colleagues performed serial biopsies on 103 patients at a mean interval of 3.2 years.[11] The fibrosis stage progressed in 37% of patients. In a similar study, Ekstedt and colleagues found that progression of liver fibrosis occurred in 41% of patients.[12]

To our knowledge, this case is the first report of NASH associated with type III cryoglobulinemia. In this patient, an extensive work-up was negative for chronic inflammatory states except for NASH. This case is significant because liver disease associated with cryoglobulinemia is largely due to chronic HCV infection. Steatosis and fibrosis have been independently associated with cryoglobulinemia but typically in the presence of HCV infection.

Saadoun and colleagues published a study of 437 patients with untreated HCV infection who had been admitted for liver biopsy.[13] Of those enrolled, 286 patients had cryoglobulins, and 103 patients had evidence of vasculitis on physical examination. Of the 286 patients in whom serum cryoglobulins were present, 186 had evidence of steatosis of greater-than-10% on liver biopsy. Multivariate analysis suggested a 3-fold increased risk of steatosis in patients with cryoglobulins, suggesting an independent association between cryoglobulinemia and steatosis. While these patients were HCV-positive, the increased risk of cryoglobulinemia with steatosis was statistically significant, and the association was interpreted as being independent of HCV infection.

In cases of cryoglobulinemia in patients who are HCV-negative, evaluation of other causes of liver disease, particularly NASH, is worthwhile. Future studies will need to elucidate a possible relationship between NASH and cryoglobulinemia independent of HCV infection.

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Review

Cryoglobulins in Nonalcoholic Fatty-Liver Disease: What Is the Association?

Benjamin C. Bryson, MD

Paul Angulo, MD, AGAF, FACG

Division of Digestive Diseases & Nutrition
University of Kentucky Medical Center
Lexington, Kentucky

The history of cryoglobulinemia dates back to the 1930s, when it was first described in a patient with multiple myeloma.[1] The modern era of cryoglobulinemia dates to 1966, when it was described by Meltzer and colleagues in a group of 9 patients presenting with a clinical triad of purpura, arthralgias, and weakness.[2] Mixed cryoglobulinemia has been better characterized since 1990, after the discovery of the hepatitis C virus (HCV).[3]

Address correspondence to:
Dr. Paul Angulo, Division of Digestive Diseases & Nutrition, University of Kentucky Medical Center, 800 Rose Street, Room MN 649, Lexington, KY 40536; Tel: 859-323-3555; Fax: 859-257-8860; E-mail: paul.angulo@uky.edu

The exact pathogenesis of cryoglobulinemia is unclear. It is hypothesized that cryoglobulinemia is the result of a disturbed immune cascade with elevated B-cell activity, and the condition often progresses smoothly to non-Hodgkin lymphoma.[4,5] Mixed cryoglobulinemias are associated with chronic inflammatory states such as systemic lupus erythematosus, Sjögren syndrome, and viral infections, particularly HCV infection.[3,6] The variable manifestations of cryoglobulinemia are due to ischemia of tissues caused by occlusion of the vessel lumen. Symptoms, when present, range from a mixed cryoglobulin syndrome (eg, purpura, arthralgias, and asthenia) to more serious systemic vasculitis with neurologic and/or renal involvement. The prevalence of symptomatic cryoglobulinemia is estimated at 1:100,000. However, the overall prevalence of patients with circulating cryoglobulins is difficult to ascertain and is probably underestimated due to the overall prevalence of HCV infection worldwide.

Studies have well established that the prevalence of nonalcoholic fatty-liver disease (NAFLD) in developed countries is increasing, paralleling the increasing incidence of obesity and metabolic syndrome.[7] NAFLD has become the most prevalent cause of chronic liver disease worldwide. NAFLD can affect individuals of any age, race, and/or ethnicity. NAFLD is estimated to affect 1 in 3 adults, resulting in over 70 million adult Americans with this condition. Additionally, it is believed that 1 in 10 children/adolescents in the United States are affected by NAFLD.[7]

The case reported by Giangreco and colleagues presents a 44-year-old male patient with a history of hypertension, hyperlipidemia, and fasting glucose intolerance who initially presented with elevated liver enzyme levels and a 6-week history of an erythematous macular/papular rash involving the trunk with sparring of the palms and soles.[8] This presentation was associated with fever, chills, and arthralgias. Initially, the patient was diagnosed with contact dermatitis and treated with steroids. Subsequently, the patient had total resolution of symptoms for 6 weeks. However, symptoms then recurred, again associated with myalgias and fever.

At that time, laboratory evaluation with regard to elevated liver enzyme levels revealed a largely negative serologic work-up for chronic liver disease, including negative results for serum viral hepatitis testing. Serologic work-up for hemochromatosis, Wilson disease, α-1 antitrypsin deficiency, and autoimmune hepatitis was negative, and HCV RNA was undetectable. Rheumatologic serologic work-up was also conducted and revealed a positive rheumatoid factor (RF; 51 IU/mL), low-titer positive antinuclear antibody (ANA) and anti–smooth muscle antibody at 1:40, decreased complement 4 levels, and normal complement 3 levels. Results of an extractable nuclear antigen panel—including ribonucleoprotein-smith, anti–Sjögren syndrome A, anti–Sjögren syndrome B, SCL-70, and anti–Jo-1—were negative, as were peripheral antineutrophil cytoplasmic antibody testing results and anticardiolipins. Immunoglobulins, light chains, and serum protein electrophoresis were normal. Serum tested positive for the presence of cryoglobulins with a pattern consistent with type III cryoglobulinemia.

A biopsy of the left thigh revealed superficial acute leukocytoclastic vasculitis. A liver biopsy revealed mixed microvesicular and macrovesicular steatosis involving 80% of the hepatic parenchyma with bridging fibrosis, mild portal chronic inflammation, and balloon-cell changes consistent with nonalcoholic steatohepatitis (NASH). Symptoms responded to immunosuppression but recurred 1 year after self-discontinuation of treatment. Rash responded again with reinitiation of mycophenolate mofetil.

What Is the Association Between Cryoglobulinemia and Nonalcoholic Steatohepatitis?

The most simplistic explanation for the association between the 2 conditions in the case reported by Giangreco and colleagues is that, since NAFLD is so common in the general population, it would not be surprising to have cases of NAFLD coexisting with essentially any other medical condition, including cryoglobulinemia.[8] However, before we assume that this explanation is correct, there are several observations that should be made regarding the case by Giangreco and associates.[8]

The prevalence of cryoglobulins in chronic liver disease was evaluated in a series of 226 patients with a wide range of chronic liver diseases.[9] Ninety-four of the 226 patients (41.6%) had cryoglobulins. Of the 127 patients with chronic HCV infection, cryoglobulins were found in 69 patients (54%), frequently with anti-HCV antibodies and HCV RNA concentrated in the cryoprecipitates. Type II cryoglobulins were present in 22 patients, and type III cryoglobulins were present in 47 patients. Eighteen of these patients had clinical symptoms compatible with cryoglobulinemia. In addition to the HCV-positive patients in this series, 40 patients had chronic hepatitis B virus (HBV) infection, and 59 patients had other hepatic diseases. Cryoglobulins were found in 15% of HBV-positive patients, 32% of patients with other liver diseases (including 45% of patients with alcoholic hepatitis or cirrhosis, 18% of patients with non-A/B/C chronic hepatitis, 33% of patients with primary biliary cirrhosis, and 50% of patients with autoimmune hepatitis), and in none of the 2 patients with steatosis. The prevalence of cryoglobulinemia in each of these groups was enhanced with increasing duration of disease and the presence of cirrhosis.[9]

To our knowledge, no prior case reports have documented the association of NAFLD and cryoglobulinemia. As mentioned previously, mixed cryoglobulinemia has been most frequently described in association with chronic HCV infection, but it can also be related to a variety of other inflammatory states. The case presented by Giangreco and colleagues meets diagnostic criteria for advanced NASH, and the presence of bridging fibrosis is worrisome, as it indicates a long duration of liver disease and a high risk of progression to cirrhosis and end-stage liver disease.[8,10] Interestingly, more than 70% of the liver parenchyma was infiltrated by fat, and both steatosis and fibrosis have been independently associated with cryoglobulinemia in the setting of chronic HCV infection.[6]

A low-titer positive ANA at 1:40 was found in this patient concurrent with a strongly positive RF test result (51 IU/mL) and decreased complement 4 levels, all of which seem to fit the clinical picture of mixed cryoglobulinemia. However, epidemiologic studies have found that healthy, asymptomatic individuals can also have positive ANA titers (25–30% at 1:40, 10–15% at 1:80, and 5% at 1:160), with a prevalence that increases with age.[10] RF is an antibody (most commonly an immunoglobulin M antibody) that is directed against the constant fragment portion of the immunoglobulin G protein. RF can be present in many conditions, including rheumatoid arthritis (RA; 26–90%), mixed connective tissue disease (50–60%), mixed cryoglobulinemia (40–100%), Sjögren syndrome (75–95%), and systemic lupus erythematosus (20–30%); RF can also be present in normal individuals (5–10%).

Like the results of ANA testing, the results of RF testing must be interpreted carefully, paying particular attention to both clinical symptoms and titers. In patients with early undifferentiated arthritis, high-titer RF (>50 IU/mL) can differentiate RA from other disorders with a specificity of 91–96%, but high-titer RF suffers from low sensitivity (45–54%).[11] Thus, a clinician could entertain the possibility of subclinical RA or mixed connective tissue disease in the setting of concurrent NAFLD, which might account for the picture of mixed cryoglobulinemia. Giangreco and coauthors did not mention musculoskeletal physical examination findings or radiologic data regarding the joints in question that might have confirmed the presence of cartilaginous changes.[8] Additionally, no information was given with regard to the nature of the patient's arthralgias. Prior authors have proposed that cryoglobulins may be the consequence of chronic liver disease secondary to decreased immune complex clearing; however, supporting data are limited.[12]

Giangreco and coauthors appropriately mention the study by Saadoun and coauthors that reported a 3-fold increased risk of steatosis in patients with cryoglobulins and chronic HCV infection.[6,8] However, it is difficult to relate the causality of steatosis to cryoglobulinemia in these patients, as the entire cohort was infected with HCV. Since the prevalence of NAFLD in the general population is high, it is reasonable to say that several of the patients may also have had an overlapping component of NAFLD.

Finally, HCV antibodies and HCV RNA were undetectable in this case. In a 1994 study, Davis and colleagues reported that there was progressive and significant loss of HCV RNA activity in the first-generation polymerase chain reaction (PCR) technique when the duration from the formation of the clot until centrifugation was longer than 2 hours.[13] Currently, HCV PCR assays have the ability to detect HCV RNA at a level as low as 10 IU/mL, so it would be unlikely that the single HCV RNA test result reported by Giangreco and colleagues was a false-negative.[8] Nonetheless, it would be interesting to see if repeat HCV testing of whole blood and serum cryoglobulins would again yield a negative result.

The interesting case reported by Giangreco and associates will likely prompt investigators to determine the prevalence of cryoglobulins in groups of patients with well-characterized NAFLD.[8] If this prevalence is higher than the prevalence of cryoglobulins in a well-matched control group, then further studies would be warranted to determine any shared mechanistic pathways between cryoglobulinemia and the development and severity of NASH.

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