Brief Communication: Case Reports of Ribavirin Treatment for Chronic Hepatitis E

Vincent Mallet, MD, PhD; Elisabeth Nicand, MD; Philippe Sultanik, MD; Catherine Chakvetadze, MD; Sophie Tessé, PhD; Eric Thervet, MD, PhD; Luc Mouthon, MD, PhD; Philippe Sogni, MD, PhD; and Stanislas Pol, MD, PhD

Background: There is currently no accepted treatment of chronic hepatitis E virus (HEV) infection.

Objective: To report 2 patients in whom ribavirin therapy seemed to alter the natural history of chronic HEV infection.

Design: Case reports.

Setting: Hepatology unit of a tertiary care center in France.

Patients: A kidney and pancreas transplant recipient and a patient with idiopathic CD4+ T lymphocytopenia, both with biopsy-proven chronic HEV infection.

Intervention: Patients received oral ribavirin, 12 mg/kg of body weight daily for 12 weeks.

Measurements: Liver function tests, detection of HEV RNA (viremia and stool shedding) by reverse transcriptase polymerase chain reaction, and anti-HEV IgM and IgG antibodies.

Results: Both patients had normalized liver function test results after 2 weeks of treatment and cleared HEV after 4 weeks of treatment. Hepatitis E virus RNA remained undetectable in the serum and stools throughout follow-up (3 months and 2 months for the first and second patient, respectively). Side effects were considered mild.

Limitation: Given the relatively short follow-up, the achievement of HEV eradication could not be claimed.

Conclusion: Ribavirin is a potentially effective treatment of HEV infection and should be evaluated in patients with chronic HEV infection.

Primary Funding Source: None.


Hepatitis E virus (HEV) is highly endemic in developing countries and is an emerging autochthonous (locally acquired) disease in industrialized countries in which a preferential zoonotic method of transmission is reported (1). Acute HEV infection has a high mortality rate in older persons, pregnant women, and patients with underlying chronic liver disease (2–5). Acute HEV infection can also evolve into chronic infection in immunocompromised patients (6, 7). The reported rate of chronicity after HEV infection is approximately 60% in solid-organ transplant recipients with persistent viral shedding observed in blood, stool, and liver biopsy samples for up to 7 years (8–10).

The diagnosis of HEV infection is based on the detection of HEV RNA in blood, stool, or liver biopsy samples and specific anti-HEV IgM and IgG antibodies. In some patients, including nonimmunocompromised patients, the course, maturation, and duration of anti-HEV IgG and IgM antibodies may be altered, and silent serologic acute or chronic hepatitis E cases have been reported (7, 9).

Chronic HEV infection can lead to cirrhosis and end-stage liver disease; in transplant recipients, it can lead to loss of liver graft (8, 11, 12). The prevalence of chronic HEV infection in transplant recipients is estimated from 0.4% in low endemic areas to 1.9% in areas with a higher prevalence (8, 12, 13). The incidence of chronic HEV infection in immunocompromised patients is difficult to estimate because of a lack of systematic HEV screening in this population. Nevertheless, the French National Reference Laboratory has reported an increasing number of cases of chronic HEV infection in immunocompromised patients in France (14).

Currently, no accepted treatment of chronic HEV infection exists, although cases of HEV clearance have been reported in transplant recipients once their immunosuppressive medication was tapered or when interferon-α was administered. Both approaches, however, introduce the risk for graft rejection (10, 15, 16). Regarding management of acute HEV infection, clinical intervention is limited to supportive care.

Ribavirin [1-(β-D-ribofuranosyl)-1,2,4-triazole-3-carboxamide] is a guanosine analogue that has broad-spectrum activity against DNA and RNA viruses, including activity against Orthomyxoviridae, Herpesviridae, respiratory syncytial viruses, Flaviviridae, and agents of many viral hemorrhagic fevers (17). Ribavirin (combined with interferon) has been used since the early 1990s to treat chronic hepatitis C infection and has been used as monotherapy in transplant recipients with chronic hepatitis C virus infection (18–21). We report the efficacy of ribavirin in 2 immunocompromised patients with chronic HEV infection.

See also:
Print
Editors’ Notes .............................................. 86
Web-Only
Conversion of graphics into slides
**Context**

Chronic hepatitis E virus (HEV) infection is an emerging infectious disease in immunocompromised patients. Therapy has been limited to supportive measures.

**Contribution**

In 2 patients with chronic HEV infection who received treatment with ribavirin, liver enzyme levels normalized and the virus was no longer detected in blood and stool. These results persisted after treatment was stopped.

**Caution**

Longer follow-up of these 2 patients is needed. Controlled trials of ribavirin treatment in patients with chronic HEV infection are needed to determine efficacy. In addition, ribavirin is contraindicated in pregnancy (U.S. Food and Drug Administration category X drug).

**Implication**

Antiviral therapy of chronic HEV infection may be possible.

—The Editors

**CASE REPORTS**

**Case 1**

The first patient was a diabetic white man aged 40 years who, after 2 years of hemodialysis, received a first simultaneous kidney and pancreas transplant from a deceased donor in November 1998. The immunosuppressive regimen included an initial 10-day course of antithymocyte globulins; corticosteroids, 500 mg/d at day 0 followed by rapid tapering to reach 10 mg/d; tacrolimus dosed to achieve a targeted trough concentration between 10 and 15 ng/mL; and mycophenolate mofetil, 1000 mg/d, adapted to clinical outcome. The patient did not have any signs of acute rejection and was free of insulin when discharged, with a serum creatinine level of 90 μmol/L (1 mg/dL).

In January 2006, the patient presented to our unit for unexplained hepatitis lasting longer than the previous 38 months. Physical examination was normal. Serum alanine aminotransferase (ALT) level was 1.6 times the upper limit of the normal range. Aspartate aminotransferase (AST), γ-glutamyltransferase (GGT), and alkaline phosphatase levels were normal. Total bilirubin level was 19 μmol/L (1.1 mg/dL), with predominance of unconjugated bilirubin. Serum creatinine level was 144 μmol/L (1.6 mg/dL). Prothrombin time was normal. The patient tested positive for anti–hepatitis C virus antibodies and negative for anti–hepatitis B virus IgG core antibodies. Genome amplifications of hepatitis B virus, hepatitis C virus, cytomegalovirus, herpes simplex virus, and Epstein–Barr virus were all negative. Ferritin and transferrin saturation blood levels were normal; antiliver and antinuclear antibodies were negative; and serum α1-antitrypsin, copper, and ceruloplasmin levels were normal—which ruled out hemochromatosis, autoimmune hepatitis, and α1-antitrypsin deficiency and Wilson disease, respectively. Liver ultrasonography findings were normal. Hepatitis C virus genome remained undetectable over time (limit of detection, 12 IU/mL).

Hepatitis E virus infection was diagnosed in September 2008 on the basis of amplification of HEV RNA (genotype 3f; GenBank accession number GU936617) and detection of anti-HEV IgG and anti-HEV IgM antibodies (EIAgen HEV IgG and EIAgen HEV IgM, Adaltis, Bologna, Italy) in the serum (Figure 1). In our study, HEV RNA was amplified with primers targeting the HEV open reading frame 2 region. Two reverse transcriptase polymerase chain reaction (RT-PCR) methods were used: real-time polymerase chain reaction that was sensitive for all HEV genotypes and conventional RT-PCR, followed by sequence analysis of the 330-nucleotide open reading frame 2 product. The detection limit of RT-PCR was 100 copies of equivalent genome per milliliter of serum by using serial 10-fold dilutions of plasmid DNA (pGEM-HEV) as a standard curve (22). Amplification of glyceraldehyde 3-phosphate dehydrogenase was used as an internal control to rule out RT-PCR inhibitors. An IgG avidity index at 97% suggested past contact with the virus. Serum and plasma samples collected at the time of the transplantation in 1998 and stored until 2008 at −80 °C were screened retrospectively, and possible exposure to the virus was dated to 2003 based on the detection of both anti-HEV IgM and IgG antibodies; however, HEV RNA could not be amplified in both techniques. In 2007, HEV RNA was detected, showing a 99.5% identity with the genome sequence amplified in 2008. A 30-mm liver biopsy (spanning 10 portal tracts) was done in April 2009 and showed evidence of chronic hepatitis with moderate activity and extensive fibrosis (METAVIR activity [A] and fibrosis [F] scores of A2 and F3). The liver biopsy showed no steatosis, steatohepatitis, or vascular abnormality. The CD4+ T-lymphocyte count was 0.72 × 109 cells/L at that time. The CD8+ T-lymphocyte count was 0.58 × 109 cells/L. The patient had never traveled to endemic areas but did report frequent consumption of undercooked pork, suggesting an autochthonous zoonotic transmission method of HEV.

In September 2009 (82 months after the presumed date of initial infection), ribavirin treatment, 400 mg twice daily (total daily dose, 12.3 mg/kg of body weight), was initiated with the informed consent of the patient. The immunosuppressive regimen was not modified. Two weeks later, ALT, AST, and GGT levels returned to within-normal limits. Four weeks after the initiation of ribavirin treatment, HEV RNA was undetectable (under the limit of detection of the assay) in the serum, and HEV IgM antibodies were negative. No fecal specimen was available for HEV RNA detection at that time. All of these findings were confirmed at weeks 6, 8, 10, and 12 of treatment. Ribavirin dose was progressively decreased to 200 mg/d because of mild anemia (nadir hemoglobin level, 92 g/L), and treatment was stopped after 12 weeks. Three months...
Figure 1. Laboratory data over 2 years for a kidney and pancreas transplant recipient with chronic HEV infection treated with oral ribavirin for 12 wk.

The graph plots the number of times the ALT level was above the ULN, by date. Genome amplifications of hepatitis B virus and hepatitis C virus were both negative. Other causes of chronic liver disease, including hemochromatosis, autoimmune hepatitis, \( \alpha_1 \)-antitrypsin deficiency, and Wilson disease, were also ruled out. For the serologic and RT-PCR tests, a plus symbol indicates a positive result and a minus symbol indicates a negative result. ALT = alanine aminotransferase; ELISA = enzyme-linked immunosorbent assay; HEV = hepatitis E virus; RT-PCR = reverse transcriptase polymerase chain reaction; ULN = upper limit of normal.

The second patient was a white woman aged 57 years with idiopathic CD4+ T lymphocytopenia and a primary deficiency in IgG-1, -2, and -4 subclasses. Her medical history included recurrent upper respiratory tract infections since childhood, with secondary bronchiectasis and moderate chronic respiratory insufficiency, and chronic cutaneous and genital human papillomavirus infection that evolved to chronic epidermodysplasia verruciformis (since age 7 years), Bowen disease (age 47 years), spinocellular cancer (age 47 years), and intraepithelial neoplasia of the vulva (age 48 years). She also reported primary cancer of the breast (age 37 years), appendicular abscess (age 52 years), and several episodes of urolithiasis.

She was first seen in our unit in July 2009 for abnormal liver function test results and a 10-kg weight loss over the previous 6 months. Apart from the dermatologic lesions, the physical examination was normal. Levels of ALT, AST, GGT, and alkaline phosphatase were 8, 5, 7.5, and 1 times the upper limit of the normal range, respectively. Total bilirubin level was 14 \( \mu \text{mol/L} \) (0.8 mg/dL). The CD4+ T-lymphocyte count was 0.22 \( \times 10^9 \) cells/L. The CD8+ T-lymphocyte count was 0.05 \( \times 10^9 \) cells/L. The CD19+ B-lymphocyte count was 0.03 \( \times 10^9 \) cells/L. The serum creatinine level was 63 \( \mu \text{mol/L} \) (0.7 mg/dL). Prothrombin time index was 97%. The patient tested negative for anti–hepatitis A virus IgM antibodies, anti–hepatitis C virus antibodies, anti–hepatitis B virus IgG core antibodies, and anti–HEV antibodies (HEV IgG enzyme-linked immunosorbent assay, Abbott Laboratories, Park, Illinois). Genome amplifications of hepatitis B virus, hepatitis C virus, cytomegalovirus, herpes simplex virus, and Epstein–Barr virus were negative. Ferritin blood level was normal; antiliver and antinuclear antibodies were negative; and serum \( \alpha_1 \)-antitrypsin, copper, and ceruloplasmin levels were normal. Liver ultrasonography findings were normal.

In December 2009, a 9-mm liver biopsy (spanning 10 portal tracts) showed evidence of chronic hepatitis, with moderate activity and mild fibrosis (METAVIR scores of A2 and F1). At that time, HEV RNA was amplified (genotype 3c; GenBank accession number HM066937) from both serum and stool samples (Figure 2), and a low anti-HEV IgM reactivity (ELAgen HEV IgM) was detected in the absence of a detectable anti-HEV IgG antibody (ELAgen HEV IgG). Two previous serum samples collected in April and July 2009 (stored at \(-80^\circ\)C) were positive for HEV RNA and anti-HEV IgM antibody, confirming chronic HEV infection. The patient reported having traveled to the south of France in November 2008 where she consumed pig liver sausage, suggesting autochthonous zoonotic transmission. Two weeks later (12 months after the presumed date of infection), ribavirin treatment, 600 mg/d divided into 2 doses (total daily dose, 12 mg/kg), was initiated with the informed consent of the patient. Two weeks later, ALT and AST levels returned to normal.
limits. After 4 weeks of treatment, HEV RNA was undetectable in the serum and stools. Ribavirin treatment was stopped after 12 weeks. Two months after the end of treatment, liver function test results remained normal and HEV RNA was not amplified from either serum or stool.

**DISCUSSION**

Because ribavirin monotherapy has broad-spectrum antiviral activity against RNA viruses and is rather well tolerated, we investigated its efficacy in 2 immunocompromised patients with biopsy-proven chronic HEV infection lasting for 82 months and more than 12 months, respectively. We observed a normalization of liver function test results after 2 weeks of treatment, and clearance of HEV RNA was attained by week 4 in both patients. To our knowledge, this is the first description of the efficacy of a nucleoside analogue for the management of patients with hepatitis E. Given the absence of available treatment, our results suggest that ribavirin should be tested in larger patient cohorts as a strategy for improving the natural course and management of patients with severe forms of HEV. One limitation of this case report is the lack of long-term follow-up. Clearly, viral eradication is the desired outcome, and evaluation of different dosing and treatment schedules may be required to achieve this goal. Nonetheless, our findings represent the first milestone toward an effective treatment of chronic HEV infection without increasing the risk for graft rejection in transplant recipients or increasing the level of primary or acquired immune deficiency.

Ribavirin is phosphorylated in human cells to ribavirin monophosphate, which is then converted to ribavirin triphosphate. Ribavirin monophosphate inhibits inosine monophosphate dehydrogenase and consequently reduces cellular guanosine triphosphate levels, which can interfere with viral (and host) nucleic acid synthesis. Ribavirin triphosphate directly interacts with the viral polymerase. We observed the same kinetics of normalization of liver function test results and of HEV clearance in both patients—1 of them having received mycophenolate mofetil, which is another inosine monophosphate dehydrogenase inhibitor. This observation could favor a direct inhibition of HEV polymerase by ribavirin rather than an antiviral activity on the basis of guanosine depletion. This hypothesis should be evaluated in vitro (23).

The primary toxicity of ribavirin is anemia. When ribavirin dose is based on weight (800 to 1400 mg/d) and is used in combination with interferon, the reported rate of anemia (hemoglobin level <100 g/L) is 19.3%. Anemia induced by ribavirin is usually mild and may be reversed...
when dosing is decreased. In the management of chronic hepatitis C virus, this approach obviates the need to interrupt treatment (24). Ribavirin-associated anemia usually occurs during the first 2 weeks of treatment, and the maximum decrease in hemoglobin levels is typically observed during the first 8 weeks. Therefore, close monitoring of hemoglobin levels is mandatory.

The U.S. Food and Drug Administration classifies ribavirin as a category X drug, indicating high teratogenic probability based on animal studies and defined mechanism of action. Hence, the drug is contraindicated in pregnant women. Ribavirin should not be used in pregnant patients with HEV infection because of the preliminary nature of these 2 case reports (25).

In conclusion, ribavirin seems to have potent activity against HEV and should be further evaluated in chronic as well as severe forms of acute HEV infection.

From Institut Cochin, Université Paris Descartes (Unité Mixte de Recherche S1016), Centre National de la Recherche Scientifique (Unité Mixte de Recherche 8104), Institut National de la Santé et de la Recherche Médicale U1016, Centre Universitaire des Saints-Pères (Unité Mixte de Recherche 775), Assistance Publique-Hôpitaux de Paris, Groupe Hospitalier Cochin Saint-Vincent de Paul, Hôpital Necker-Enfants Malades, and Hôpital d’Instruction des Armées du Val-de-Grâce, Paris, France.

Note: Drs. Mallet, Sogni, and Pol contributed equally to this article.

Acknowledgment: The authors acknowledge the patients’ readiness to help others by accepting their case be the focus of this article. The authors also thank Anais Vallet-Pichard, MD; Hélène Fontaine, MD; Matthew Albert, MD, PhD; and Marie François Avril, MD, PhD, for their help in the care of the reported patients and for their advice on the manuscript.

Potential Conflicts of Interest: Disclosures can be viewed at www.acponline.org/authors/icmje/ConflictOfInterestForms.do?msNum=M10-0831.

Reproducible Research Statement: Study protocol and statistical code: Not available. Data set: Available from Dr. Mallet (e-mail, vincent.mallet@cc.aphp.fr) after establishing written agreement with the authors.

Requests for Single Reprints: Vincent Mallet, MD, PhD, Assistance Publique-Hôpitaux de Paris, Groupe Hospitalier Cochin Saint-Vincent de Paul, Unité d’Hépatothérapie, 27 rue du Faubourg Saint Jacques, 75014 Paris, France; e-mail, vincent.mallet@cc.aphp.fr.

Current author addresses and author contributions are available at www.annals.org.

References