Two Successive Hepatitis C Virus Infections in an Intravenous Drug User

BARBARA PROUST,1 FREDERIC DUBOIS,1,YANNICK BACQ,2 SOPHIE LE POGAM,1 SYLVIE ROGEZ,3 ROMUALD LEVILLAIN,1 AND ALAIN GOUDEAU1

Département de Microbiologie Médicale et Moléculaire, Unité de Virologie, Centre Hospitalier Universitaire Bretonneau,1 and Service d’Hépatogastroentérologie, Centre Hospitalier Universitaire Trousseau,2 Tours, and Laboratoire de Bactériologie-Virologie-Hygiène, Centre Hospitalier Universitaire Dupuytren, Limoges,3 France

Received 8 February 2000/Returned for modification 18 April 2000/Accepted 6 June 2000

We report the case of an occasional intravenous drug user who developed two successive hepatitis C virus (HCV) infections. The first infection led to seroconversion (anti-HCV antibodies detected) and the detection of HCV RNA in serum. After a spontaneous recovery (normalization of alanine aminotransferase levels and HCV RNA clearance), a second HCV infection was observed, with the recurrence of HCV viremia. Antibody directed against HCV serotype 1 was detected throughout the follow-up monitoring, but two different HCV strains were identified during the two infectious episodes: genotype 1a for the first and genotype 3a for the second. This observation shows that primary HCV infection does not confer protective immunity against subsequent infection with viruses of other genotypes. This may hamper the development of a vaccine. Conflicting results were obtained in genotyping and serotyping assays, suggesting that the serotyping method cannot be used to identify the HCV type in patients, such as intravenous drug users, who are exposed to successive HCV infections.

CASE REPORT

A 30-year-old woman was referred to a hepatology unit in January 1996 with a diagnosis of acute hepatitis, which started in December 1995 with asthenia, nausea, and icterus. Tests carried out at the onset of infection showed a rise of alanine aminotransferase (ALT) levels (1,996 IU/liter) and the presence of antibody against hepatitis C virus (anti-HCV), detected with a third-generation enzyme-linked immunosorbent assay (ELISA) (ORTHO HCV 3.0 ELISA Test System with Enhanced SAVe; Ortho-Clinical Diagnostics, Neckargemund, Germany). This patient reported occasionally injecting drugs intravenously and sharing needles with a single female friend in October 1995. She was not infected by the human immunodeficiency virus (HIV) (anti-HIV negative; anti-HBs, >500 IU/liter). On admission, the presence of anti-HCV was confirmed by both ELISA and a third-generation recombinant immunoblot assay (RIBA) (Chiron RIBA HCV 3.0 SIA; Ortho Diagnostic Systems). HCV RNA was detected by reverse transcription (RT)-PCR (Amplicor HCV Test; Roche Diagnostic Systems, Neuilly sur Seine, France). The virus was identified as genotype 1a by a second-generation line probe assay (INNO-LIPA HCV II; Innogenetics, Ghent, Belgium) and as HCV serotype 1 by a determination of type-specific antibodies against NS4-derived peptide antigens (Murex HCV serotyping assay, version 1-6; Murex Diagnostics, Chatillon, France) (Table 1). The notion that the HCV infection was recent was based on negative results of a test for HCV in April 1995, when the patient gave a blood donation, seroconversion (appearance of anti-HCV antibodies), and the increase in levels of anti-HCV antibodies detected by RIBA between December 1995 and March 1996 (Table 1). The patient did not receive antiviral treatment.

Follow-up monitoring suggested a full recovery, as shown by the disappearance of clinical symptoms, the normalization of ALT levels, the disappearance of HCV RNA from serum, and the decrease in titer of antibodies against NS5 (Table 1).

In March 1998, this patient again consulted doctors for asthenia and pain in the right hypochondrium. Because she was pregnant, she was tested for antibody to rubella (result, >200 IU/ml), and her serological status with regard to HBV was controlled (results, HBsAg negative; anti-HBs, >500 IU/liter). She tested negative for human immunodeficiency virus and did not receive medical treatment. She had abnormal liver biochemistry results, in particular a slight increase in ALT levels (65 IU/liter). She had again used intravenous drugs in August and September 1997 and had shared needles with a single female friend different from the one involved in the 1995-1996 episode. Antibodies against NS5 and HCV RNA were again detected (Table 1). HCV genotype 3a was identified during this second HCV infection. Surprisingly, serotyping showed antibody to serotype 1 for all samples collected during both episodes. No reaction with serotype 3 was observed at any time during follow-up monitoring (Table 1). The second HCV infection was not resolved after nine months of monitoring.

Discussion. The proportion of spontaneous resolution of an HCV infection following acute hepatitis C is estimated to be 30% (4). Recovery of this type is suggested by the disappearance of HCV-RNA viremia and the return to normal of ALT levels. However, there are no specific criteria for determining whether patients who recover in this way are protected against subsequent infections. It has been reported that spontaneous viral clearance may be related to host immunity, with a mechanism involving a human leukocyte antigen class II-restricted T lymphocyte response to a nonstructural viral protein (5). Thus, the host human leukocyte antigen genetic factor may influence the ability to overcome HCV infection (1, 13, 17). Few data are available concerning the immune response to HCV infection, although highly conserved and immunogenic sequences have been identified in the viral core, NS3 and NS4 proteins (10).
has been shown that the protective immunity developed after experimental HCV infection in chimpanzees is relatively weak and does not protect against reinfection with homologous or heterologous HCV strains (6, 19).

Our case shows that the first HCV infection with a genotype 1a virus did not confer protective immunity against subsequent infection with a heterologous genotype 3a strain. It demonstrates that HCV reinfection is possible in a patient who fully recovered from a first HCV infection with HCV-RNA clearance and normalization of ALT levels. Rare cases of HCV reinfection have been reported for patients who received several transfusions (9). One case has also recently been reported for an intravenous drug user (16). Thus, the possible lack of induction of protective immunity by HCV infection, the heterogeneous nature of the virus, and its ability to mutate rapidly appear to prevent the development of an effective neutralizing immune response, hampering the development of an effective vaccine.

We cannot fully rule out a double infection in 1996, with a predominance of type 1a during the first years and emergence of the second genotype during the 1998 outbreak. Such an event has never been reported in the literature. Our patient reported sharing needles exclusively with two different female friends during the 1995-1996 and 1997-1998 episodes, and we hypothesize that they were the source of the two successive infections.

The results of HCV serotyping and genotyping tests were not consistent at the time of the second HCV infection. The serotyping assay falsely indicated infection with serotype 1 during the second HCV episode, although the HCV strain responsible was of genotype 3. This observation may be explained by the "original antigenic sin" phenomenon originally described for influenza vaccination (2, 3). In essence, it appears that immunization with antigen α (here HCV type 1) can establish a population of memory B cells such that subsequent infection with a related antigen β (here HCV type 3) stimulates a high-affinity response to the antigen α but a lower-affinity response to the antigen β. In this study, infection with the second HCV strain presumably boosted the specific antibody response to the original. Although it cross-reacted with the new strain, a higher-affinity response for the original HCV strain would thus explain the predominance of antibodies to serotype 1 during the second HCV episode. Similar phenomena have been reported for other diseases, such as dengue virus infection, and have been used to identify the initial serotype of a previous infection so as to clarify the antecedents of dengue shock syndrome (7, 8).

Previous studies have suggested that the sensitivity of HCV to interferon alfa differs according to HCV genotype, with patients infected with HCV genotype 3 having a sustained and better response to interferon alfa therapy than patients infected with genotype 1 (14, 15, 18). The use of less-costly serotyping assays for the identification of HCV isolates in the clinical setting to guide treatment decisions has been proposed (11, 12). In our case, serotyping results during the second infection episode were misleading. Thus, deduction of the HCV genotype from serotyping results could give false results for subjects, such as intravenous drug users, who are exposed to the risk of successive infections.

This clinical case demonstrates that protective immunity may not develop in cases of HCV infection, leading to the possibility of HCV reinfection. During reinfection, serotyping may be of value for identifying virus strains involved in previous episodes, whereas the genotyping of current isolates should be the method of choice for guiding treatment decisions.

REFERENCES


TABLE 1. Successful HCV infections: changes in serological markers

<table>
<thead>
<tr>
<th>Date of testing (day/mo/yr)</th>
<th>ALT levels (IU/liter)a</th>
<th>Assay for HCV antibody by:</th>
<th>HCV serotype</th>
<th>HCV-RNA analysis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>ELISA-b</td>
<td>RIBA-3 for antibody againstc</td>
<td>Genotype</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Core NS3 NS4 NS5</td>
<td></td>
<td></td>
</tr>
<tr>
<td>First infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>21/12/95b</td>
<td>1,996</td>
<td>+ ++ + + + + + + + + + + + +</td>
<td>ND</td>
<td>ND</td>
</tr>
<tr>
<td>03/01/96</td>
<td>265</td>
<td>+ + ++ + + + + + + + + + + +</td>
<td>1</td>
<td>1a</td>
</tr>
<tr>
<td>07/02/96</td>
<td>31</td>
<td>+ + ++ + + + + + + + + + + +</td>
<td>1</td>
<td>1a</td>
</tr>
<tr>
<td>25/03/96</td>
<td>21</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>1a</td>
</tr>
<tr>
<td>21/08/96</td>
<td>7</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>1a</td>
</tr>
<tr>
<td>14/09/98</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Second infection</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>19/03/98b</td>
<td>65</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>3a</td>
</tr>
<tr>
<td>15/06/96</td>
<td>44</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>3a</td>
</tr>
<tr>
<td>14/09/98</td>
<td>19</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>3a</td>
</tr>
<tr>
<td>05/12/96b</td>
<td>23</td>
<td>+ ++ ++ + + + + + + + + + + +</td>
<td>1</td>
<td>3a</td>
</tr>
</tbody>
</table>

a The upper limit of normal ALT levels was 35 IU/liter.
b In all cases, the ratio of optical density to the cutoff was >4.
c RIBA-3 reactivities were rated from (−) to +4 according to the intensity of the bands.
d Retrospective analysis was carried out by a neighboring laboratory. ND, not done. No samples were available with which to carry out other HCV analyses (serotype, genotype, and RT-PCR) retrospectively.
e First month of pregnancy.
f Delivery.