and a response to specific anti-PCP therapy. All 83 cases are included in the following analysis. The number of cases was evenly distributed over the 4 years of study, but after 1990 there was a significant increase in the number of patients in whom primary PCP developed while they were receiving prophylaxis. Overall, 69 (83%) patients were homosexual males. The mean (± SD) CD4 cell count within 6 months of the diagnosis of PCP was 60 ± 88/mm³ (range, 0–500/mm³ for 63 patients). Five of the 63 patients had CD4 cell counts of >200/mm³, but in all five cases the percentage of CD4 cells was <20 (range, 9%–19%). None of the five patients were receiving prophylaxis when PCP was diagnosed.

Sixty-seven patients were being observed by physicians when PCP was diagnosed. Most of these physicians were expert in the care of HIV-infected individuals. However, only 21 patients were receiving prophylaxis for PCP when this infection was diagnosed (16 were receiving pentamidine, 4 were receiving dapsone, and 1 was receiving trimethoprim-sulfamethoxazole [TMP-SMZ]). Since prophylactic pentamidine was administered in our clinic, we could make a precise estimate of compliance based on patients’ records. Except for two patients who were receiving dapsone therapy, all patients who were receiving prophylaxis were deemed compliant.

We explored the reasons why 62 patients were not receiving prophylaxis at the time of primary PCP. Eighteen had previously tried preventive therapy but discontinued it; 12 discontinued therapy because of an allergy to TMP-SMZ or dapsone, 2 because of intolerance to other medication, 3 by patient choice, and 1 because of disease progression. Only 6 of these 18 patients had tried and discontinued all approved prophylactic medications. Twenty patients had not sought medical care for HIV infection before presenting with PCP. They were more likely to have been immigrants (P < .001) and/or women (P < .01) and to have acquired HIV heterosexually (P < .01) when compared with those who were observed by physicians. We could not find a clear reason why 28 patients were not receiving prophylaxis for PCP.

Hence, despite the availability of prophylactic medications and clear guidelines for their use, preventing primary PCP remains difficult. PCP continued to occur in our cases because of the failure of physicians to provide continuous prophylaxis for those at risk, the failure of the prophylactic medications themselves (due to either toxicity [TMP-SMZ] or poor efficacy [pentamidine]), and the failure of patients to seek timely medical care (particularly immigrants and those who acquired HIV heterosexually). Steps towards further decreasing the incidence of PCP should include physician and patient education, public health measures that are culturally sensitive to nontraditional risk groups, and the development of more-effective and better-tolerated prophylactic medications.

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References

Fatal Hepatic Necrosis in a Neonate with Echovirus 20 Infection: Use of the Polymerase Chain Reaction to Detect Enterovirus in Liver Tissue

Enteroviruses have been shown to cause severe infections in newborns [1], and hepatitis is one of the most severe manifestations of this infection. We report a case of fatal hepatic necrosis associated with echovirus 20 infection that occurred in a newborn during an outbreak of echovirus 20 infection in France in 1994. This serotype has never been implicated as a cause of severe perinatal disease. Only 6 of these 18 patients had tried and discontinued all approved prophylactic medications. Sixteen patients who were receiving prophylaxis were deemed compliant.

Except for two patients who were receiving dapsone therapy, all patients who were receiving prophylaxis were deemed compliant. We explored the reasons why 62 patients were not receiving prophylaxis at the time of primary PCP. Eighteen had previously tried preventive therapy but discontinued it; 12 discontinued therapy because of an allergy to TMP-SMZ or dapsone, 2 because of intolerance to other medication, 3 by patient choice, and 1 because of disease progression. Only 6 of these 18 patients had tried and discontinued all approved prophylactic medications. Twenty patients had not sought medical care for HIV infection before presenting with PCP. They were more likely to have been immigrants (P < .001) and/or women (P < .01) and to have acquired HIV heterosexually (P < .01) when compared with those who were observed by physicians. We could not find a clear reason why 28 patients were not receiving prophylaxis for PCP.

Hence, despite the availability of prophylactic medications and clear guidelines for their use, preventing primary PCP remains difficult. PCP continued to occur in our cases because of the failure of physicians to provide continuous prophylaxis for those at risk, the failure of the prophylactic medications themselves (due to either toxicity [TMP-SMZ] or poor efficacy [pentamidine]), and the failure of patients to seek timely medical care (particularly immigrants and those who acquired HIV heterosexually). Steps towards further decreasing the incidence of PCP should include physician and patient education, public health measures that are culturally sensitive to nontraditional risk groups, and the development of more-effective and better-tolerated prophylactic medications.

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References

A 3,010-g infant girl was born on 25 July 1994 to a 24-year-old primigravida by cesarean section because of cephalopelvic dystocia. The mother became febrile at the time of delivery and was treated with ampicillin. The infant had an Apgar score of 9 at 1 minute after birth and appeared normal. On the fourth after birth, the infant suddenly became febrile (temperature of 38.2°C) and jaundiced and was immediately transferred to the pediatric department. Bacteriological cultures of blood, urine, and CSF were negative, and pleocytosis was not evident. Therapy with cefotaxime and amikacin was initiated. The infant’s condition deteriorated rapidly. Her clinical symptoms included hepatosplenomegaly, jaundice, and pulmonary and gastric hemorrhage. The child was intubated.

Laboratory tests revealed a platelet count of 26,000 cells/mm³ and a prothrombin time of 11.9 seconds (16%; normal, >40%–100%). Her partial thromboplastin time was 120 seconds, and her fibrinogen level was 57 mg/dL. Fibrin split products were 40 mg/L (normal, <10 mg/L). Coagulation factors were below normal: factor V was 5% (normal, 83%–105%), factor VII plus X was 3% (normal, 10%–63%), and factor II was 4% (normal, 23%–85%). The concentrations of aspartate aminotransferase and alanine aminotransferase in serum were 3,710 U/L and 460 U/L, respectively (normal, <40 U/L).
The infant underwent exchange transfusion, but her condition deteriorated. The results of coagulation studies were still abnormal; she continued to bleed, and thrombocytopenia persisted despite multiple platelet transfusions. She died 10 days after birth.

Bacteriological cultures of blood, urine, and CSF remained negative. There was no serological evidence of infection due to hepatitis A, B, or C; cytomegalovirus; rubella; adenovirus; or Toxoplasma gondii. Serology for HIV was negative.

A urine specimen that was obtained for viral culture on the fourth day after birth was inoculated into human lung fibroblast MRC5 cell lines. Enterovirus was rapidly isolated, and the virus was identified as echovirus 20 with use of Lim-Benyesh-Melnick antiserum pools. No specific IgM and IgG antibodies to echovirus 20 were detected in the neonate. However, specific antibody titers of IgM (1:80) and IgG (1:1,024) to echovirus 20 were detected in serum specimens from the mother 10 days after birth; the same level of these titers was detected in a third serum specimen obtained on August 26, 22 days after the baby had died. No virological samples were obtained at delivery.

An autopsy did not reveal congenital anomalies or evidence of myocarditis. Samples from the brain showed cytoplasmic swelling without necrosis and vacuolation of white matter. Samples from the liver showed macroscopic and histological features of shock (i.e., massive liver cell necrosis). Only one or two layers of cells situated around the portal area and centrilobular vein were spared. Consequently, a diagnosis of generalized hepatic necrosis was made. Samples from the liver were frozen at -80°C for further investigations.

Enterovirus genome was detected after cDNA synthesis by a nested PCR assay of a hepatic biopsy specimen. The PCR assay amplifies a 435-bp segment of the 5'-noncoding region of the genome. The nucleotide sequence of the primers is indicated in Table 1. The first PCR assay was performed with the primer pair 5NC63 and 5NC642. No specific product was visualized by agarose gel electrophoresis. However, nested PCR amplification of a hepatic tissue specimen that was performed with the primer pair 5NC615 and 5NC599, resulted in a 435-bp product, whereas no product was amplified with use of the negative controls.

To our knowledge, we report the first case in which echovirus 20 infection has been associated with hepatic necrosis and in which the enterovirus genome has been detected with use of nested PCR of hepatic fetal tissues. Hepatic necrosis has been associated with echoviruses 6, 7, 9, 11, 14, 19, and 21 (mainly with serotype 11) and with coxsackievirus B [1, 4]. Echovirus 20 has already been implicated in neonatal infection, but only in inapparent infection [1]. Eichenwald et al. recovered echovirus 20 from the nasopharynxes of four full-term apparently well infants who were <8 days old [5]. Cesarean section may have increased our patient's risk of infection by interrupting the pregnancy before the maternal specific IgG antibody was transferred.

Since the first description of echovirus 20 in 1958, it has only rarely been implicated in outbreaks of disease [6]. We believe that, since echovirus 20 has not been previously reported as a cause of massive hepatic necrosis, the impact of genetic variations in echoviruses should be considered [7-9]. At present, more than 10% of wild enterovirus strains isolated worldwide fail to type with reference immune sera [9]. Such collected strains should be investigated at a molecular level, and Romero et al. [10] have recently begun such studies of neuroviral echovirus types.

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**References**


**Table 1. Oligonucleotide primers used in the nested PCR assay of a hepatic biopsy specimen.**

<table>
<thead>
<tr>
<th>Oligonucleotide</th>
<th>Sequence</th>
<th>Reference</th>
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<tbody>
<tr>
<td>5NC63</td>
<td>5'-GGTACCTTTTGGCCCCCTG-3'</td>
<td></td>
</tr>
<tr>
<td>5NC642*</td>
<td>5'-CACCGGATGCGCAATCCA-3'</td>
<td>[2]</td>
</tr>
<tr>
<td>5NC165*</td>
<td>5'-CAGCCTTCTGTTCCCCCGG-3'</td>
<td>[3]</td>
</tr>
<tr>
<td>5NC599*</td>
<td>5'-ATGTCACCAAAGCCAGCCA-3'</td>
<td>[3]</td>
</tr>
</tbody>
</table>

* Called primer D in [2].
† Oligonucleotides 5NC165 and 5NC599 were called primer 1 and primer 3, respectively, in [3].