Frequent Occurrence of Chronic Hepatitis B Virus Infection among West African HIV Type-1–Infected Children

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Background. The aim of this study, conducted in Ivory Coast, was to evaluate the prevalence and evolution of viral hepatitis in children coinfected with human immunodeficiency virus type 1 (HIV-1).

Methods. Hepatitis B virus (HBV) and hepatitis C virus (HCV) markers were retrospectively and longitudinally assessed among 280 HIV-1–infected children enrolled in the Agence Nationale de Recherches sur le SIDA et les Hépatites Virales B et C 1244/1278 cohort. Among these, 173 (61.8%) received highly active antiretroviral therapy (HAART), including lamivudine (3TC) for 122 children. Detection of the hepatitis B s antigen (HBsAg) was performed on specimens collected at inclusion and 6 months later. If results of both tests were positive, hepatitis B e antigen (HBeAg)/hepatitis B e antibody (HBeAb) and HBV DNA levels were measured at inclusion and during follow-up. A fourth-generation HCV enzyme immunoassay was used for HCV screening at inclusion.

Results. In our pediatric cohort, no patients were infected with HCV, but the prevalence of HBsAg at inclusion was 12.1% (34 of 280; 95% confidence interval [CI], 8.6–16.6). Among the HBV–HIV-1–coinfected children, a high rate of positive HBeAg chronic hepatitis B (CHB) was noted at inclusion (82.4% [28 of 34]; 95% CI, 65.5%–93.2%) and after a median follow-up of 18 months (78.3%; 95% CI, 45.5%–92.7%), with no significant difference between children treated with HAART (with or without 3TC) and untreated ones. These children showed high HBV DNA levels (usually >8.0 log10 copies/mL) and viral population consisting of nearly exclusively wild-type HBeAg-positive HBV strains, strongly suggesting that most of them were in the initial immunotolerant phase of chronic hepatitis B.

Conclusion. In sub-Saharan Africa, children with chronic hepatitis B and who are treated with 3TC-based HAART are at risk of developing 3TC resistance. Further studies are required to guide the management of HBV–HIV-1–coinfected children.

Worldwide, 387 million people are estimated to be chronic carriers of hepatitis B virus (HBV), which is the main cause of chronic liver disease (e.g., cirrhosis and hepatocellular carcinoma) and liver-related mortality. In Africa, the number of HBV carriers is estimated to be ~50 million [1]. Many African countries where HBV prevalence is high (≥8%) are also affected by HIV-1 infection, yielding frequent HBV–HIV coinfections. The number of pediatric HBV–HIV coinfections must be high, because most HBV infections occur within the first 5 years of life, and mother-to-child transmission of HIV-1 is an important public health problem.

In developed countries, with the widespread use of HAART, the clinical impact of HBV–HIV-1 coinfections is quite apparent, since risks of mortality from
classic opportunistic infections have diminished tremendously [2, 3]. Because treatment with HAART is scaling up in many African countries, it is likely that liver disease due to chronic hepatitis B (CHB) will also emerge in Africa [4]. Unfortunately, data are scarce on HBV–HIV-1 coinfections in Africa [5], notably among children [6, 7].

At present, 7 drugs are available for treating CHB [8–11]. Among these, 4 drugs (i.e., lamivudine [3TC], tenofovir [TDF], emtricitabine [FTC], and entecavir [ETV]) possess a dual (anti-HBV and anti-HIV) activity. 3TC is the most widely available drug throughout Africa. Hepatitis B e antigen (HBeAg) seroconversion has been reported in 22%–29% of HBV–HIV–coinfected adult patients who receive 3TC, whereas undetectable HBV DNA levels were achieved in 40%–87% of patients [12–19]. However, 3TC monotherapy leads inevitably to selection in a high proportion of resistant HBV strains, with mutations of the YMDD motif in the catalytic or C domain of HBV polymerase gene [8, 20]. Simultaneous therapy with 3TC and TDF has a more beneficial effect on coinfected adults than does 3TC or TDF added to 3TC [21, 22].

The main objectives of the present study, conducted among HIV-1–infected children enrolled in the Agence Nationale de Recherches sur le SIDA et les Hépatites Virales B et C (ANRS) 1244/1278 cohort in Abidjan, Ivory Coast, were (1) to determine the prevalence of hepatitis B and C and (2) to study the HBeAg seroconversion rates among children with CHB, regardless of whether they have been treated with HAART (with or without 3TC), through a 30-month follow-up period.

**PATIENTS AND METHODS**

**Pediatric population.** The studied population consisted of HIV-1–infected children enrolled from October 2000 until December 2003 in the ANRS 1244/1278 observational children cohort [23, 24]. The ANRS 1244/1278 protocol had been approved by the National Health Authorities and the National Ethics Committee of Ivory Coast. At baseline, each child underwent a clinical and psychological evaluation. Clinical evaluations were performed quarterly during the follow-up period. Children were also seen for any intercurrent diseases, if necessary. All diseases and health-related events during the follow-up period were recorded on a standardized form. Consultations at the outpatient clinic and the day hospital, treatments, and tests were free of charge to the patient.

**Laboratory methods.** The detection of HBs antigen (HBsAg) was determined on samples obtained at inclusion and 6 months thereafter by using the Murex HBs Ag version II EIA (Abbott Laboratories). CHB was defined as a positive HBsAg result at inclusion in the cohort and was confirmed at 6-month follow-up, if samples were available. All available samples from inclusion to 30 months of follow-up obtained from children exhibiting a CHB were further tested for ongoing viral repli-

cation with use of HBeAg/AntiHBe (Murex HBeAg/AntiHBe EIA; Abbott Laboratories). When available, these samples were also tested for HBV DNA (Quantiplex HBV DNA; Bayer Diagnostics). The detection ranges of the branched DNA assay were 3.3–8.0 log_{10} copies/mL of HBV DNA.

Antibodies to hepatitis C virus (HCV) were screened on samples taken at inclusion with use of a fourth-generation (G4) EIA (Innotest HCV Ab IV; Innogenetics). Specimens with a signal-cutoff ratio <1.0 were considered to be negative, whereas those with a signal-cutoff ratio ≥4.0 were considered to be positive. All specimens that were positive, with a signal-cutoff ratio range of 1.0–4.0, were further assessed with a G3 EIA (Ortho HCV 3.0 ELISA; Ortho-Clinical Diagnostics) [25].

CD4+ T cell counts and plasma HIV-1 RNA load measurements were performed at baseline and every 6 months thereafter. Percentages of CD4+ T cells were measured by flow cytometry (FACScan; Becton Dickinson). Plasma HIV-1 RNA levels were initially determined using the Versant HIV-1 RNA version 3.0 assay (Bayer Diagnostics). The detection threshold of this assay was 250 copies/mL (i.e., 2.4 log_{10} copies/mL), determined by use of 0.2 mL of plasma. Since April 2003, HIV-1 RNA load measurements were determined using a real-time RT-PCR test targeted in the HIV-1 LTR gene. The detection threshold of this assay was 300 copies/mL (i.e., 2.5 log_{10} copies/mL) with use of 0.2 mL of plasma [26].

**Statistical analysis.** HBV DNA and HIV-1 RNA were log_{10} transformed for analysis. For prevalence of HBV and HCV infection, exact 95% CIs were calculated. The Kaplan-Meier method was used to estimate the probability of HBeAg persistence among children with CHB.

**RESULTS**

**HBV and HCV infection prevalence at inclusion.** The main baseline characteristics of the 280 HIV-1–infected children included in the ANRS 1244/1278 cohort are summarized in table 1. One hundred seventy-three (61.8%) of the 280 children were treated with HAART during follow-up. Most of them (112 [64.7%] of 173) began treatment after their inclusion in the cohort. One hundred twenty-two received a 3TC-containing HAART regimen (4 mg/kg twice daily).

HBsAg was detected at inclusion in 34 children, yielding a CHB prevalence rate of 12.1% (34 of 280; 95% CI, 8.6%–16.6%) (table 1). The presence of HBsAg was confirmed among 25 children whose blood could be tested on samples obtained at month 6. The 9 remaining children died (n = 6) or were lost to follow-up (n = 3) before month 6. Six of the 9 were not treated with HAART at inclusion. No significant difference was observed between these 34 children who were HBsAg positive at inclusion and those who remained HBsAg negative, in terms of sex, age, median percentages of CD4+ T cells, and HIV-1 RNA values. Centers for Disease Control and Prevention
stages B and C were significantly more frequent in HBsAg-positive children than they were in those who were HBsAg negative (29 of 34 vs. 156 of 246; \(P = .01\), by \(\chi^2\) test). Seven children who were HBsAg positive died. The causes of death were cachexia \((n = 2)\), tuberculosis \((n = 2)\), pneumonia \((n = 2)\), and unknown \((n = 1)\).

Among the 34 HBsAg-positive children, 28 were also HBeAg positive (including 26 children without and 2 with detectable HBeAb) (figure 1). Thus, the prevalence of HBeAg-positive CHB was 82.4% \((28 \text{ of } 34; 95\% \text{ CI}, 65.5\%–93.2\%)\). Additionally, 1 child was HBeAg negative/HBeAb positive, whereas 5 were HBeAg negative/HBeAb negative. Among these 34 children, 23 could also be tested for HBV DNA at inclusion. All but 1 child with HBeAg-positive CHB had a high titer of HBV DNA copies \((\text{median}, >8.0 \log_{10} \text{copies/mL})\). One child who was HBeAg negative/HBeAb negative had a positive HBV DNA test result \((4.48 \log_{10} \text{copies/mL})\).

Of 280 HIV-1–infected children, 278 were found, using the Ingen EIA, to be negative for anti-HCV antibodies. Two samples initially found to be weakly positive (signal-cutoff ratio range, 1–4) with this technique were strictly negative with the Ortho EIA. Thus, the anti-HCV prevalence was 0% \((95\% \text{ CI}, 0.0\%–1.3\%)\) (table 1).

**Longitudinal follow-up of CHB in HIV-1–coinfected children treated with HAART.** Of the 25 HBsAg-positive children who could be followed up (median duration of follow-up, 18 months; interquartile range, 18–30 months), 16 children were eligible for and were treated with first-line HAART regimens. Two of them (patients 014 and 106) switched their regimens during follow-up.

Eleven children received a 3TC-containing HAART regimen. The evolution of their HBV and HIV-1 markers was as follows:

1. 4 children exhibited good HBV and HIV-1 virological responses—for instance, children 134 and 138 (figure 2A and 2B) had high HBV DNA levels \( (>8.0 \log_{10} \text{copies/mL}) \) at inclusion and exhibited undetectable HBV DNA results at the end of their follow-up;
2. 3 children exhibited low response rates against both HBV and HIV-1—for instance, children 014 and 106 (figure 2C and 2D) showed high \( (>8.0 \log_{10} \text{copies/mL}) \) HBV DNA levels at each time point;
3. 3 children showed a low HBV-response rate, on the basis of positive HBeAg at each time, but a good immunovirological response against HIV-1; and
4. 1 child showed undetectable HBV DNA results but a virological HIV-1 RNA failure.

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**Table 1. Baseline hepatitis B virus and hepatitis C virus status among 280 Ivorian HIV-1–infected children.**

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Findings</th>
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<tbody>
<tr>
<td>Female sex</td>
<td>128 (45.7)</td>
</tr>
<tr>
<td>Age, median years (range)</td>
<td>5.8 (1.4–15.8)</td>
</tr>
<tr>
<td>Received HAART during follow-up</td>
<td>173 (61.8)</td>
</tr>
<tr>
<td>Received HAART containing 3TC during follow-up</td>
<td>122 (43.6)</td>
</tr>
<tr>
<td>Clinical stage</td>
<td></td>
</tr>
<tr>
<td>N</td>
<td>8 (2.8)</td>
</tr>
<tr>
<td>A</td>
<td>87 (31.1)</td>
</tr>
<tr>
<td>B</td>
<td>133 (47.5)</td>
</tr>
<tr>
<td>C</td>
<td>52 (18.6)</td>
</tr>
<tr>
<td>Median CD4+ T cell count, % (range)</td>
<td>14.4 (0.1–43.7)</td>
</tr>
<tr>
<td>Median plasma HIV-1 RNA load, log_{10} copies/mL (range)</td>
<td>5.02 (Undetectable–7.34)</td>
</tr>
<tr>
<td>Positive for hepatitis B s virus antigen(^a)</td>
<td>34 (12.1)</td>
</tr>
<tr>
<td>Positive for hepatitis C virus antibody</td>
<td>0 (0.0)</td>
</tr>
</tbody>
</table>

\(\text{NOTE.}\) Data are no. (%) of subjects, unless otherwise indicated. All percentages are of the cohort of 280 children. Subjects are from the Agence Nationale de Recherches sur le SIDA et les Hepatites Virales B et C 1244/1278 cohort, Abidjan, Ivory Coast, 2000–2004. 3TC, lamivudine.

\(^a\) The presence of hepatitis B’s antigen was confirmed at month 6 among 25 children.
On the other hand, 7 children were treated with HAART without 3TC. All but 1 showed high HBV DNA levels during their entire follow-up periods. Two had good HIV-1 immunovirological responses, whereas 5 showed virological HIV-1 RNA failure.

**Longitudinal follow-up of CHB in untreated HIV-1–coinfected children.** Seven of 9 children showed high HBV DNA levels ($>8.0 \log_{10}$ copies/mL) during their entire follow-up periods, as shown in figure 2E for child 141. One child (087) experienced a spontaneous HBeAg seroconversion at month 18 (figure 2F). One child exhibited a similar pattern, but his HBV DNA level was not assessed at his last time point.

**Probability of persistent positive HBeAg CHB.** With use of a Kaplan-Meier analysis (figure 3), among the 21 children who experienced HBeAg-positive CHB at inclusion and who could be followed up, the overall probability of persistent HBeAg-positive CHB was 100% at month 6, 100% at month 12, and 78.3% (95% CI, 45.5%–92.7%) at month 18. No significant difference was observed between children treated with HAART (with or without 3TC) and untreated children.

**DISCUSSION**

Our study, conducted in Ivory Coast, showed that HBV coinfection is endemic among HIV-1–infected children with an HBsAg prevalence rate >10%. The prevalence of HBeAg-positive CHB is also high in these immunodepressed HBV-HIV–coinfected children, with a probability of HBeAg persistence of
as part of HAART, it must be continued indefinitely. Indeed, options later in life [30, 31]. Additionally, once 3TC is started, the body knows whether 3TC resistance can lead to fewer treatment regimens when considering management of CHB in HBV–HIV-1–coinfected children. In fact, specific guidelines are very difficult to establish. The complexity of HBV, HIV, and HAART interactions must be evaluated for each individual. The optimal time to initiate anti-HBV treatment in HIV-1–coinfected subjects is not established, especially among children. As reported in a study conducted in Thailand [27], the immunotolerant phase can last for decades and is usually associated with a very low risk (<0.5% per year) of developing cirrhosis or hepatocellular carcinoma. Thus, the substantial benefit from specific HBV treatment remains unknown.

In resource-constrained countries, for children who have reached clinical and/or immunological criteria for anti-HIV therapy, HAART without 3TC should be the best option. However, first-line HAART regimens very often contain 3TC. Second-line anti-HIV regimens are still rare and expensive. To date, for anti-HIV treatment, given the lack of alternative regimens, 3TC-based HAART constitutes the only available option for these patients. Physicians must be aware that 3TC-based HAART can induce a high rate (~20% per year) of HBV resistance in HIV-1–coinfected patients [28, 29]. However, nobody knows whether 3TC resistance can lead to fewer treatment options later in life [30, 31]. Additionally, once 3TC is started as part of HAART, it must be continued indefinitely. Indeed, if coinfected subjects are switched to a second-line HAART regimen, discontinuation of 3TC can result in HBV reactivation and flares in the alanine transaminase level [20, 32]. Finally, in sub-Saharan Africa, the challenge of treating CHB specifically in HIV-1–coinfected children is currently quite difficult. HBV-specific drugs (such as adefovir, telbivudine, and pegylated IFN-α-2a) are not available. 3TC is associated with a high risk of selection of resistant HBV strains. TDF is not licensed for children. Other agents with anti-HBV activity (such as FTC and ETV) are not available.

Our study has several limitations: no conclusion about the natural history of HBV-HIV coinfection in childhood could be done, because main complications of HBV infection (i.e., cirrhosis and hepatocellular carcinoma) are observed only many years after infection [33]. In fact, at present, the long-term evolution of CHB in African HBV–HIV-1–coinfected children remains unknown. The relatively small sample size and the lack of liver enzyme measurements are other limitations of our study.

In conclusion, our study showed that CHB was frequent in our population of HIV-1–infected children. In resource-limited settings where HAART is scaling up, testing for CHB must be implemented. Indeed, coinfected subjects have more cirrhosis and increased toxicity from HAART and are at a greater risk of liver-related death [10, 34]. At the least, presence of HBsAg and liver enzyme levels must be determined before starting HAART. When possible, the detection of HBeAg and HBV DNA levels will be also helpful to determine whether there is presence of active HBV replication and, thus, the need (or not) for anti-HBV therapy. For this purpose, low-cost, homemade real-time PCR assays for HBV DNA quantitation [35] should be evaluated in Africa. Further studies are clearly needed to create new guidelines for managing the increasing number of children who are being treated with HAART and have CHB. To face this growing public health problem, the introduction of the HBV vaccine at or soon after birth into the Expanded Program of Immunization, as done in Ivory Coast since June 2002, appears also as a major method for preventing HBV infection in this sub-Saharan country. HBV revaccination among treated HIV-1–infected children with immune recovery after HAART must also be urgently considered [36].

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