Hepatitis B virus prevalence and vaccination response in health care workers and students at the Federal University of Bahia, Brazil

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ABSTRACT

Background and rationale for the study. Hepatitis B (HB) is one of the most prevalent occupational infections in health attendance environments. According to the Brazil Ministry of Health, health professionals must be vaccinated against the hepatitis B virus (HBV) and provide laboratory proof of immunization. Aims. To evaluate the seroprevalence of HBV infection and to analyze the response to vaccine by measuring serum antibodies against HBV surface antigen (anti-HBs) levels in a sample of students and health professionals at the Federal University of Bahia. Results. As part of this cross-sectional study, a campaign against occupational HB was launched in 2007 and vaccination and blood samples were collected for analysis of the following serological markers: HBV surface antigen (HBsAg) and anti-HBs (measured by enzyme-linked immunoassay) and total antibodies against HBV core antigen (anti-HBc). The study sample comprised 766 people. Global seropositivity for HBV was 1.7%; 0.5% in the students and 8.8% in the professionals. In a group of volunteers, a serological profile compatible with postvaccine immunity was shown by 95% of volunteers with proof of vaccination and by 81.8% of volunteers without proof of vaccination. Conclusions. In conclusion, this study shows that it is important to promote vaccination campaigns and improve knowledge and awareness about HB among health care workers and higher education students.

Key words. Hepatitis B vaccination program. Healthcare students and professionals. Occupational hepatitis.

INTRODUCTION

Infection by the hepatitis B virus (HBV) is a main cause of hepatic disease throughout the world. It is estimated that among the two billion persons infected by the virus, there are 360 million chronic carriers of the disease.¹ Most people infected chronically by HBV present no active hepatic disease and are defined as inactive carriers. However, persistent infection may cause future cirrhosis, hepatic failure, or hepatocellular carcinoma.²-⁴ HBV has oncogenic potential and was the first virus proven to be responsible for a type of cancer in humans.⁵,⁶ Cirrhosis caused by HBV is one of the main indications for liver transplant and represents a high cost to public health.⁴

The distribution of hepatitis B is universal but the prevalence rates vary throughout the world.² In Brazil in the 1990s, three hepatitis B distribution patterns were observed: high endemicity (prevalence > 7%) in the Amazonian region, south of Espírito Santo, and in the western part of the states of Paraná.
The prevalence of HBV serological markers is higher in health professionals than in the general population. HBV is considered one of the most important infections contracted in hospital environments.\textsuperscript{9,10} The risk of contracting HBV infection is closely related to the manipulation of blood and its derivatives.\textsuperscript{11} Depending on the viremia level, the risk of viral transmission through injuries caused by contaminated objects is estimated at 6 to 30%.\textsuperscript{12} There are two types of immunobiological products to provide prophylaxis against HBV infection: anti-hepatitis B immunoglobulin, which provides temporary protection (3 to 6 months) and is indicated only after accidental exposure, and hepatitis B vaccine, which provides long-term protection against HBV infection.\textsuperscript{13} The vaccine against HBV is the most commonly used immune prophylaxis worldwide and is scheduled in the basic vaccination calendar in various countries. Although the World Health Organization has recommended vaccination against HBV since 1997, some countries do not follow this recommendation because of the high cost of the vaccine. In Brazil, the Ministry of Health has made the vaccine against HBV available free of charge to all health professionals since 1994 and to people younger than 20 years since 2001.\textsuperscript{14}

In countries where vaccination is performed on a large scale, the epidemiology of HBV infection and its morbidity and mortality have decreased dramatically.\textsuperscript{15} Measures have been proposed to minimize HBV exposure in health professionals. In 2006, the US Centers for Disease Control and Prevention showed that the incidence of HBV infection among health professionals was lower than in the general population.\textsuperscript{16} In Brazil, although efforts have been made to vaccinate health professionals, many individuals in situations of risk do not have vaccinations or do not complete the vaccination schedule.

The aims of this study were to evaluate the HBV seroprevalence and the response to vaccine by measuring serum anti-HBs levels in a sample of students and health professionals at the Federal University of Bahia (UFBA) in Salvador, Brazil, during the year 2007.

**MATERIALS AND METHODS**

A cross-sectional study was conducted with a sample comprising students and professionals in the health area of UFBA. Students came from the courses of medicine, nursing, pharmacy, biology, phonoaudiology, dentistry, biology, and nutrition, and professionals were drawn from the teaching units of the programs in medicine, nursing, pharmacy, biology, phonoaudiology, dentistry, and the multidisciplinary teaching unit (Instituto de Ciências da Saúde or Institute of Health Sciences). The following professions were included in the professional category: professors, laboratory technicians/assistants, nursing technicians/assistants, technicians, and personnel responsible for cleaning, pharmacists, doctors, nurses, physiotherapists, and speech therapists. Individuals from the administrative area, without any technical training or higher education in the areas of health, were excluded from the study.

The campaign was conducted during the year 2007 in accordance with the universal scheme of vaccination against HBV (0, 1, and 6 months) in the university health units. This was organized in four stages. During the first three stages, the volunteers completed an epidemiological and clinical questionnaire, were vaccinated, and provided a blood sample. In the fourth stage, only blood samples were collected.

The individuals with an incomplete vaccination scheme or without previous vaccination received the vaccine from the LG Life Sciences laboratory, owned by the Brazilian Ministry of Health. The vaccination scheme recommended by the National Immunization Program (Programa Nacional de Imunização) comprises three doses of vaccine, with an interval of 1 month between the first and second dose, and 5 months between the second and third dose. The vaccines were conserved and transported at a temperature of 2-8°C according to the regulations of the Brazilian Ministry of Health and were injected intramuscularly in the deltoid region by nurses. The standard vaccine concentration was 10 μg mL\textsuperscript{-1}.\textsuperscript{17}

The volunteers were classified into two groups depending on their vaccination status. Group 1 included those who showed proof of vaccination before the campaign by a vaccination card this card is proved that the vaccine status was completed in the public politics healths and the individuals that took the 3 doses of vaccine during the study; and group 2, included those who said that their vaccination...
scheme was complete, but that they did not have a vaccination card-(auto-referred status).

In the group who took the vaccination during the study, one month after the last dose of vaccine, blood samples were collected for analysis to provide a serological profile by measuring the levels of anti-HBc total, HBsAg, and anti-HBs. The tests were performed using an automated system and Immulite 2000 commercial kits in the Immulite 2000 appliance, HBs MicroEIA in the ARIO RADIM appliance, and IEMA WELL in the ALISEI RADIM appliance, Radim Latin America Diagnostic, São Paulo, Brazil.

The volunteers were classified into three profiles according to the serological results.

- **Profile 1**—immunity conferred by natural infection: anti-HBs positive ($\geq 10 \text{ mUI mL}^{-1}$) + anti-HBc-total positive + HBsAg negative.
- **Profile 2**—immunity conferred by vaccination: anti-HBs positive + anti-HBc-total negative + HBsAg negative.
- **Profile 3**—susceptible to HBV: anti-HBs negative (< $10 \text{ mUI mL}^{-1}$) + anti-HBc-total negative + HBsAg negative.

Individuals who were anti-HBc positive were referred to the Gastro-Hepatology Unit of the University Hospital Professor Edgar Santos–UFBA to complete a clinical evaluation. Polymerase chain reaction analysis (Amplicor, Roche, Basel) was used to analyze samples from individuals with profile 1 (total isolated anti-HBc positive).

For statistical evaluation of the data, with reference to the serological response to vaccine, individuals with profile 1, immunity by natural infection, were excluded. These individuals were considered immune protected with a titration of antibodies anti-HBs $\geq 10 \text{ mUI mL}^{-1}$ in accordance with the World Health Organization recommendation.\textsuperscript{2,18} Individuals with an anti-HBs antibodies level < $10 \text{ mUI mL}^{-1}$ were referred to the Reference Center for Specials Immunobiologics–UFBA for revaccination in accordance with the Brazil Ministry of Health recommendation.\textsuperscript{19}

The study was performed according to the principles of the Declaration of Helsinki and was approved by the Research Ethics Committee of the Bahia Foundation for the Development of Sciences, Report Nos. 20/2004 and 36/2004. Written informed consent was obtained in every case.

This was a descriptive study. The prevalence ratios (PRs) and their respective 95% confidence intervals (95% CI) were calculated. The $\chi^2$ test was applied to test the associations between dichotomous variables and Fisher’s exact test was used when appropriate. The results were considered significant for $p < 0.05$ or when the null value was outside of the 95% CI.

**RESULTS**

From a total population of 4,000 individuals, 766 volunteers who participate in the study had blood samples collected. The blood samples were 85.1% (652/766) from students and 14.9% (114/766) from professionals. The sociodemographic data are shown in the table below (Table 1).

About 90% (690/766) of the sera collected were from individuals with proven complete vaccination (group 1); 9.9% (76/766) did not have proof of complete vaccination (group 2). For profile 1, the prevalence of previous contact with HBV was 1.7% (13/766); these individuals were anti-HBs positive, anti-HBc-total positive, and HBsAg negative. Eleven of these 13 individuals were from group 1 and two were from group 2. Three of the individuals with profile 1 [0.5% (3/652) were students and 8.8% (10/114) were professionals ($PR = 17.61, p < 0.000, \chi^2 = 32.27$)]. The HBV DNA detected by polymerase chain reaction was negative in samples from these 13 volunteers.

Because the 13 volunteers with profile 1 had immunity conferred by previous contact with the virus, they were excluded from further statistical analysis. This left a sample of 753 volunteers who presented with profile 2 or 3: 679/753 were in group 1 (594 students and 85 professionals) and 74/753 were in group 2 (55 students and 19 professionals). The results of the analysis of serological responses to vaccination are shown in table 2.

Analyses of the response to vaccines by occupation are shown in tables 3 and 4.

Susceptibility and professional activity correlated positively ($PR = 2.70, 95\% \text{ CI 1.46-5.01}$). In group 1, the PR was 2.22 (95\% CI 0.98-5.05), and in group 2, the PR was 2.17 (95\% CI 0.86-5.45).

In group 1, 164 individuals received three doses of vaccine only during the campaign. Six volunteers were excluded from the analysis of the response to vaccine because they presented with profile 1. In this group, a total of 158 sera were analyzed, and 98.7% (156/158) showed vaccine-induced immunity. The age of these individuals was $27.14 \pm 10.7$ years. The two volunteers who had susceptibility to HBV (profile 3) were older, 37 and 53 years.
Table 1. Epidemiological profile of participants in the campaign.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Occupation</th>
<th>Student (n = 652)</th>
<th>Professional (n = 114)</th>
<th>Total (n = 766)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n (%)</td>
<td>n (%)</td>
<td>n (%)</td>
<td></td>
</tr>
<tr>
<td>• Sex</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>444 (68.1)</td>
<td>83 (72.8)</td>
<td>527 (68.8)</td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>208 (31.9)</td>
<td>31 (27.2)</td>
<td>239 (31.2)</td>
<td></td>
</tr>
<tr>
<td>• Drawn from</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Medicine</td>
<td>184 (28.2)</td>
<td>7 (6.1)</td>
<td>191 (24.9)</td>
<td></td>
</tr>
<tr>
<td>Dentistry</td>
<td>139 (21.3)</td>
<td>22 (19.3)</td>
<td>161 (21)</td>
<td></td>
</tr>
<tr>
<td>Nursing</td>
<td>144 (22.1)</td>
<td>14 (12.3)</td>
<td>158 (20.6)</td>
<td></td>
</tr>
<tr>
<td>Pharmacy</td>
<td>116 (17.8)</td>
<td>28 (24.6)</td>
<td>144 (18.8)</td>
<td></td>
</tr>
<tr>
<td>Phononaudiology</td>
<td>36 (5.5)</td>
<td>4 (3.5)</td>
<td>40 (5.2)</td>
<td></td>
</tr>
<tr>
<td>Nutrition</td>
<td>21 (3.2)</td>
<td>1 (0.9)</td>
<td>22 (2.9)</td>
<td></td>
</tr>
<tr>
<td>Biology</td>
<td>12 (1.9)</td>
<td>- (-)</td>
<td>13 (1.7)</td>
<td></td>
</tr>
<tr>
<td>ICS</td>
<td>- (-)</td>
<td>38 (33.3)</td>
<td>38 (5)</td>
<td></td>
</tr>
</tbody>
</table>

*For this variable, because this item was incomplete in some of the questionnaires, the data for 63 volunteers were lost: 46 students and 17 professionals.

± SD. ICS: Instituto de Ciências da Saúde or Institute of Health Sciences.

Table 2. Serological analysis after vaccination of the volunteers in groups 2 and 3 who were either immune or susceptible to HBV.

<table>
<thead>
<tr>
<th>Vaccination scheme</th>
<th>Group 1</th>
<th>Group 2</th>
<th>PR</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Proven vaccination</td>
<td>Vaccination not proven</td>
<td></td>
<td></td>
</tr>
<tr>
<td>• Profile 2: immunity conferred by vaccination</td>
<td>n (%)</td>
<td>n (%)</td>
<td>PR</td>
<td>95% CI</td>
</tr>
<tr>
<td>anti-HBs (+)/ anti-HBc-total (-)/ HBsAg (-)</td>
<td>650/679 (95.7)</td>
<td>60/74 (81.1)</td>
<td>1.36</td>
<td>1.10-1.67</td>
</tr>
<tr>
<td>• Profile 3: susceptible to HBV</td>
<td>Anti-HBs (-)/ Anti-HBc-total (-)/ HBsAg (-)</td>
<td>29/679 (4.3)</td>
<td>14/74 (18.9)</td>
<td></td>
</tr>
</tbody>
</table>

PR, prevalence ratio; CI, confidence interval; anti-HBs (+), anti-HBs positive; anti-HBs (-), anti-HBs negative; anti-HBc-total (-), anti-HBc-total negative; HBsAg(-), HBsAg negative.

Table 3. Prevalence ratios and 95% confidence intervals for the association between the profile of immunity to HBV and occupation.

<table>
<thead>
<tr>
<th>Profiles</th>
<th>Profile 2 (Immune after vaccination</th>
<th>Profile 3 (Susceptible to HBV)</th>
<th>PR (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n (%)</td>
<td>n (%)</td>
<td></td>
</tr>
<tr>
<td>Student</td>
<td>619/649 (95.4)</td>
<td>30/649 (4.6)</td>
<td>2.70</td>
</tr>
<tr>
<td>Professional</td>
<td>91/104 (87.5)</td>
<td>13/104 (12.5)</td>
<td>(1.46-5.01)</td>
</tr>
<tr>
<td>Total</td>
<td>710/753 (94.3)</td>
<td>43/743 (5.7)</td>
<td></td>
</tr>
</tbody>
</table>

PR: prevalence ration. CI: confidence interval.
In group 1, 94.8% (494/521) of those who received at least one dose of vaccine outside of the campaign had vaccine-induced immunity. The seroconversion rate after the vaccine did not differ significantly between the volunteers who received three doses of vaccine only during the campaign and those who received at least one dose outside the campaign (PR = 0.98, 95% CI 0.86-1.11).

**DISCUSSION**

HBV infection is recognized as an important occupational risk.10 Thus, vaccination campaigns and education about basic measures to prevent exposure to the pathogen are universally recommended to prevent infection in health care workers.14

The preliminary data from the population-based study of the prevalence of hepatitis A, B, and C in Brazil showed seropositivity variation from 0.11 to 0.74% for HBV in the northeast and mid-west regions.20 In our sample, the rate of anti-HBc-total seropositivity was 1.7 %, which is lower than the 7.9% in the Brazilian general population21 and the 3.0% in the population of Salvador-Bahia-Brazil in the 1980s.22 A study in 2000 found 1.2% prevalence of anti-HBc in the northeast region of Brazil,23 which is similar to the rate in our study, probably because of the government vaccination measures and disclosure campaigns in the past few years.

Different studies of HBV prevalence in health professionals have shown a higher incidence of this virus in specific groups compared with the general population. A study of 152 hemodialysis professionals in Goiânia (Goiás State) reported a prevalence of 24.3% for anti-HBc and/or HBsAg.24 Another study conducted at the University Hospital of Salvador in the northeast region of Brazil in 2007 found 7.6% seropositivity for HBV (anti-HBc and/or HBsAg) in a sample of 263 health professionals (PO Marques, UFBA unpublished data).

The 8.8% of professionals who had previous contact with HBV (anti-HBc-total positive and HBsAg negative) in this study was higher than the prevalence of 5.8% (anti-HBc total positive and HBsAg negative) in a study of dentists conducted in 2005 in Bahia.25 The higher percentage exposure to HBV in the health professionals included in our study compared with the general population suggests an occupational source of HBV infection.

Quantifying the prevalence of HBV infection is important in a continental country such as Brazil, which has regions with different endemicity. Standardization of these studies with the intention to evaluate the HBV serological markers is fundamental to recognizing the different endemicity and to implementing specific prevention strategies for each region.

In this study sample, seropositivity for the virus was positively associated with professional activity. The seroprevalence for previous contact with HBV was 17.6 times higher in professionals (8.8%) than in students (0.5%). This marked difference in serological profile between these two groups can be explained by the students’ younger age, less time for contact with potentially contaminated fluids, greater stimulus to use individual protective equipment, and possible prior vaccination against HBV when it was introduced in the basic childhood vaccine calendar by the Ministry of Health in the year 2000.12,14,26

Infection by HBV is a persistent long-term disease. Some individuals with a serological profile compatible with a clinical cure (HBsAg negative and anti-HBs positive) and with blood persistent complete infectious particles (HBV DNA positive) have been reported.27 This may be explained by a progressive decline in the replication and expression of the HBV genome or by the genetic variability of HBV. In this study, nobody was at risk of HBV transmission because the virus was not detected by the polymerase chain reaction technique.

### Table 4. Analysis of response to vaccines in groups 1 and 2.

<table>
<thead>
<tr>
<th></th>
<th>Group 1</th>
<th></th>
<th>Group 2</th>
<th></th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Immune</td>
<td>Susceptible</td>
<td>PR (95% CI)</td>
<td>Immune</td>
</tr>
<tr>
<td><strong>Vaccination</strong></td>
<td>after</td>
<td>to HBV</td>
<td></td>
<td>after</td>
</tr>
<tr>
<td><strong>n (%)</strong></td>
<td>vaccine</td>
<td>n (%)</td>
<td></td>
<td>vaccine</td>
</tr>
<tr>
<td>Student</td>
<td>572 (96.3)</td>
<td>22 (3.7)</td>
<td>2.22</td>
<td>47 (85.5)</td>
</tr>
<tr>
<td>Professional</td>
<td>78 (91.8)</td>
<td>7 (8.2)</td>
<td>0.98-5.05</td>
<td>13 (68.4)</td>
</tr>
<tr>
<td>Total</td>
<td>650 (95.7)</td>
<td>29 (4.3)</td>
<td></td>
<td>60 (81.1)</td>
</tr>
</tbody>
</table>
Immunoprophylaxis through a preexposure vaccine against hepatitis B is the main preventive measure among health professionals and students. The vaccine is safe and is extremely effective: 90-95% of seroconversion in immunocompetent adults. Adverse reactions to the vaccine are rare and, if they occur, are usually mild, such as hyperemia, itching, and local edema. Vaccination must be applied by a trained professional and is indicated for all people who may be exposed to biological materials during their activities, including individuals who do not work directly with patients, for example, cleaning and support teams.

The lower seroconversion rate in response to the vaccine in health professionals than in students, despite the sample number, is consistent with the rate in a previous study of the humoral response to the vaccine, which monitored the presence of anti-HBs over three years. Age is considered an important factor affecting the serological response. Younger individuals exhibit seroconversion with higher titers of anti-HBs, and the titer level correlates inversely with age.

Gauging the effectiveness of a vaccine based on self-report may lead to an overestimation of the prevalence of vaccination because most people know the importance of the vaccine. This study confirms this concept because the group with proof of three doses of vaccination had a significantly higher seroconversion rate than did the group without proof of vaccination. In individuals who said they had been vaccinated but had no proof, the susceptibility to HBV might indicate that fewer doses had been received. Although the groups vaccinated outside of the campaign were not evaluated in this study, other conditions may cause a negative response to vaccination including the preservation method for the vaccine, age at vaccination, time that elapsed between vaccination and measuring anti-HBs, obesity, smoking, chronic diseases, and immunosuppression.

In situations in which health professionals do not display serological levels of anti-HBs ≥ 10 mUI mL⁻¹, the Brazilian Ministry of Health recommends revaccination with three doses at 0, 1, and 6 months. If the negative result persists, these individuals are considered nonresponders by the conventional triage methods available for research on anti-HBs recommended by the World Health Organization. In the event of accidental exposure to a contaminated perforating-cutting instrument, the nonresponding professional should receive a dose of hepatitis B hyperimmune immunoglobulin, which is extremely effective when applied within 28 h. The risks and benefits of maintaining these professionals in assistive activity with imminent risk of contamination by the virus should be evaluated.

In the group that received three doses of vaccine only during the campaign, the rate of seroconversion was similar to that of another study in which the same brand of vaccine was used. The two volunteers who did not seroconvert in response to the vaccination scheme were 37 and 42 years old. These data are consistent with another study that observed better response to vaccines in young individuals. The rate of seroconversion to the vaccine did not differ significantly between these individuals and those in group 1, who received at least one dose outside of the campaign. This result shows that a good response can be induced with different brands of the vaccine.

To prevent occupationally acquired infectious diseases, it is important to promote adequate preventive vaccination among health care workers who have a high risk of contamination with biological fluids. The observation that some individuals who were anti-HBs negative had been vaccinated reinforces the importance of serological evaluation after the vaccination scheme in this specific population. The presence of unvaccinated individuals and another ones who didn’t realize the study of serological response to vaccination, suggest that some people did not fully appreciate the role of the vaccine as an effective preventive measure. Further study is needed to assess the real immunoprophylaxis status of health workers in all health care institutions.

This study shows the importance of improving knowledge and awareness of hepatitis B among health care providers, especially in developing countries such as Brazil. We suggest that health care workers should consider the vaccination card an important document for recording their clinical and occupational history in different situations.

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and Molecular Biology Laboratory of the Health Science Institute (Laboratório de Imunologia e Biologia Molecular do Instituto de Ciências da Saúde).

ABBREVIATIONS

- HB: hepatitis B.
- HBV: hepatitis B virus.
- HBsAg: HBV surface antigen.
- anti-HBs: antibodies against HBsAg.
- anti-HBe: antibodies against HBV core antigen.
- UFBA: Federal University of Bahia.

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