

Short Communication

Anti-Hepatitis B Antibody Levels In Immunized Medical Students: Are They At Risk?

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Abstract

Introduction: Medical students have an occupational risk for hepatitis B (HB). This study sought to determine anti-HBs and anti-HBc IgG levels in vaccinated students, check their seroconversion, and correlate this with vaccination. **Methods:** One hundred and forty-three students' blood samples and their vaccination schedules were analyzed. **Results:** 65.7% were positive for anti-HBs; however, anti-HBs was absent in 34.3%. Only two samples were positive for anti-HBc IgG. **Conclusions:** More than 30% of students did not have minimum protective levels. Comparing HBV vaccination and anti-HBs reactivity, the majority of reactive individuals received their last dose within the past 16 years.

Keywords: Hepatitis B. HBsAg antibodies. Vaccination. Seroconversion.

Hepatitis B is a chronic, infectious liver disease caused by double-stranded DNA from the Hepadnaviridae virus¹. Since 1994, Brazilian health authorities have recommended vaccination to infants at birth, one month, and six months of age². Those adults and high-risk groups not vaccinated at birth should receive the three doses, with the minimum interval between the first and second doses being one month, and two months between the second and third doses³.

Children and young adults who receive the three recommended doses typically achieve a 90% seroconversion rate. However, only 75% of adults gain protection against infection if they complete immunization later in life³. Levels greater than 10 mIU/ml of antibodies against the surface viral antigen (anti-HBs) are considered to be protective for human beings. In Brazil, seroconversion rates tend to be lower in all groups after full immunization. Osti and Marcondes-Machado found non-seroconversion in 18.3% of hospital workers⁴.

A study of children and adolescents showed that 50.1% had undetectable anti-HBs levels (below 10 mIU/ml)⁵.

In China, studies found high levels of anti-HBs seroconversion. Among college students, they found that only 0.8 to 1% that did not achieve anti-HBs seroprotection after complete vaccination^{6,7}. Among children and adolescents aged 5 to 15 years, the rate of non-seroconversion was lower (0.3 to 0.8%)^{7,8}. However, some factors can reduce the effectiveness of the vaccine, such as the vaccine preservation methods, age, obesity, smoking, genetic factors, and chronic diseases^{2,3}.

This cross-sectional epidemiological study sought to evaluate anti-HBs seroconversion and associated clinical factors among medical students in Southern Brazil. Currently, the University of Blumenau (FURB) Medical School has 459 students, distributed across 12 semesters. Sixty-one and seven tenths percent are female, and 38.3% are male. A convenience sampling of undergraduate medical students was recruited by institutional email. They were required to have completed the immunization schedule for hepatitis B (minimum of 3 doses) at least one month before the blood collection date and not have acute Hepatitis B at the time of collection.

From March 2015 to March 2016, students answered a structured and standardized questionnaire after signing informed

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consent. Those who had a vaccination card presented it to check the dates and number of doses. After that, the trained and qualified professionals of the Counseling and Testing Centre (CTA) collected ten milliliters of peripheral blood by venipuncture in the Laboratory of Immunology and Physiology, Department of Natural Sciences, at FURB. All samples were tested for HIV, syphilis and hepatitis B and C according CTA protocols

The dependent variables were serum anti-HBs and serum anti-HBc IgG levels. Both were determined by electrochemiluminescence immunoassay. We used specific commercial kits in a Cobas E411 device (both from Roche Diagnostics®, North America). We used the Brazilian Pediatric Society reference values for serum anti-HBs levels and the cut-off from the manufacturer for qualitative anti-HBc IgG (from 1.0 to less than 4.8: non-reactive samples, and values less than 1.0 as reactive)³. The independent variables were gender (male or female), age (in years as a continuous, grouped variable), number of years after last vaccine dose, and number of doses.

Categorical variables were presented as frequencies (absolute or relative), and numeric variables were presented as mean or median with standard deviation (SD). Numeric variables were tested for a normal distribution (Shapiro-Wilk test). We employed the Chi-square and Mann-Whitney tests for bivariate associations. We used the software Bioestat 5.3 and accepted a significance level of 0.05 for all tests.

The FURB's Ethics Committee on Human Research examined and approved this study (CAAE 35832814.3.0000.5370). All participants signed consent for the CTA to send the anti-HBs and anti-HBc IgG results to the researchers for analysis. The researchers of this study did not have access to other results, thus maintaining the anonymity provided by CTA.

The study had 143 participants, 94 (65.7%) of which were female, spread across basic training to internship. Their mean age was 22 years (SD = 3.29), median was 21, and range was 17 to 41 years. Ninety-eight individuals (68.5%) had detectable levels of anti-HBs, while 45 people (31.5%)

did not. Anti-HBc IgG was nonreactive in 141 samples and reactive in 2 cases (**Table 1**). When all samples were stratified by age, the proportion of anti-HBs positivity increased with age (**Table 1**). This indicates that there is a positive significant correlation between age categories and seroconversion. With respect to antibody titers, the mean serum level of anti-HBs in reactive individuals was 472.64 mIU/mL (median: 398, SD: 420.50), while the average anti-HBc IgG was 1.45 (median: 1.39, SD: 0.59).

Although all participants were proven to have been vaccinated against hepatitis B, the date of vaccination was not available for 33 of them. According to inoculation records, the ages at vaccination ranged from 2 to 22 years and the number of doses ranged from 3 to 8 (with only one student having had 8 doses). We found a significant relationship between the year of the last vaccine dose and serum anti-HBs levels, as shown in **Table 2**. The presence of high levels of specific antibodies is evident in students who received the last dose of vaccine more recently, compared to individuals who received it more than 16 years ago. No statistically significant correlations were found between gender and seroconversion, or between the number of vaccine doses and the levels of serum anti-HBs antibodies.

The data obtained by the anti-HBs levels showed that 45 people (31.5%) did not have the minimum levels necessary for immunization against hepatitis B virus, even after completing the full vaccination schedule. There is wide variability in the existing literature regarding the prevalence of anti-HBs levels after vaccination. In a recent study of health professionals in the southwestern United States, Bookstaver and colleagues reported 83.9% seroconversion when observed approximately 10 years after immunization⁹. In a study of the cleaning staff of the Botucatu Clinical Hospital (southeast Brazil), Osti and Marcondes-Machado, showed that 18.3% had serum anti-HBs levels lower than 10 mIU/ml⁴. According to the study by Livramento et al. that assessed children and adolescents in Blumenau, SC (southern Brazil), 39.9% of individuals were below this cut-off⁵. The results of our study are within the

TABLE 1: Epidemiological and statistical characteristics and serum levels of anti-HBs and anti-HBc IgG from students evaluated (n=143) in Blumenau/SC from February 2015 to February 2016.

Variables	Sample, n (%)	Nonreactive, n (%)	Reactive, n (%)	p
Age group				
21 or Younger	77 (53.8)	33 (23)	44 (30.8)	0.002 (#)
22 or Older	66 (46.2)	12 (8.4)	54 (37.8)	
Anti-HBs IgG		45 (31.5)	98 (68.5)	
Male	49 (34.3)	13 (28.9)	36 (36.7)	0.36
Female	94 (65.7)	32 (71.1)	62 (63.3)	
Anti-HBc IgG		141 (98.6)	2 (1.4)	

(#) chi-square test (χ^2); **HBs:** Hepatitis B surface antigen; **HBc:** Hepatitis B core; **IgG:** Immunoglobulin G.

TABLE 2: Comparison between number of years of last anti-HBV vaccination and anti-HBs levels from medical school students evaluated in Blumenau/SC from February 2015 to February 2016.

	Anti-HBs Levels				p
	Nonreactive		Reactive		
	Median	(Q1; Q3)	Median	(Q1; Q3)	
Years Post-Vaccination	20.00	(4; 21)	15.00	(3; 20)	0.039

Q1: lower quartile; **Q3:** upper quartile; **HBs:** Hepatitis B surface antigen; **HBV:** Hepatitis B virus.

prevalence margins obtained in the literature. However, it is important to note that the laboratory methods used for anti-HBs quantification differ from the literature.

Comparing age categories revealed a trend for younger individuals to have less frequent immunity (lower levels of anti-HBs antibodies), especially in those individuals younger than 22 years old. This data is in opposition to the study in Goiânia, GO (central Brazil) that showed greater likelihood of seroconversion in younger individuals¹⁰. Another relevant correlation existed between the timing since the last dose of immunization and serum anti-HBs levels (non-reactive versus reactive), as seen in **Table 2**, such that higher, reactive serum levels were found in individuals who more recently completed their last dose, which is corroborated in the literature. According to some studies, complete, post-vaccination antibody titers may decrease until they become undetectable^{2,3}. About half of young adults who had a full regimen of hepatitis B vaccination may have low or undetectable levels of anti-HBs 15 years post-vaccination. However, they may retain protection against the disease by preserving clonally-specific immune memory B and T lymphocytes^{3,11-13}. In our study, there was no association between gender and seroconversion. This conclusion agrees with the results obtained by Rezaee et al.¹⁴. However, other authors have shown higher serum levels of antibodies in females than in males⁴.

This study has some limitations. This is a convenience sample and those concerned about their immunization or serological status may have been more interested in participating. This could explain, at least in part, the high level of non-seroconversion. Further, the small sample limits the study's power and generalization. Ultimately, the percentage of students with inadequate levels of seroconversion was quite high. A recent study indicates that protection against hepatitis B depends on immune memory and not on the presence of circulating anti-HBs antibodies¹⁵. But considering the risk of contact with HBV-contaminated materials, we strongly recommend that non-seroconverted students receive a booster dose of the vaccine^{2,3}.

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