PREVALENCE OF SEROLOGICAL MARKERS FOR HEPATITIS B AND HEPATITIS C AMONG PRACTICING DENTAL HEALTHCARE PROFESSIONALS

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ABSTRACT

Background: Dental healthcare professionals (DHPs) are exposed to a significant risk of infection with blood-borne pathogens, including hepatitis B virus (HBV) and hepatitis C virus (HCV). The present study aimed to estimate the prevalence of serological markers for hepatitis B and hepatitis C among practicing dental healthcare professionals.

Material and methods: A cross-sectional survey was conducted between 1 June and 31 October 2024 to evaluate the presence of serological markers for hepatitis B and C among DHPs (N = 133). The detection of serological markers for hepatitis B and C was performed using enzyme-linked immunosorbent assay (ELISA) and chemiluminescent microparticle immunoassay (CMIA). The differences between proportions of interest were assessed. Continuous data were expressed as median with interquartile range. Numbers and percentages (n, %) were used to present qualitative variables. A z-test was conducted to evaluate disparities between proportions.

Results: The median age of the enrolled DHPs was 43 years with women outnumbering men almost fivefold. Professional accidents were self-reported by 17% of the participants as 78% were vaccinated against HBV. Dentists were the most affected. The presence of protective HBsAb was detected in 49% of the DHPs who were self-reported as vaccinated. In 37% of the enrolled DHPs, HBsAb were detected, and in 10%, concomitant HBcAb were detected, indicating a past HBV infection.

Conclusions: The findings of this study suggest a necessity for regular screening for viral hepatitis among dental professionals.

Keywords: Dental healthcare professionals, HBV, HCV

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INTRODUCTION

Dental healthcare professionals (DHPs) are at increased risk of viral infections transmitted by both airborne droplets and blood such as hepatitis B (HBV) and C (HCV) viruses. The generation of splatter and aerosols is a result of the use of ultrasonic scalers, high-speed air rotors, air-water syringes, and air polishing. At the same time, the risk of injury to both the patient and the dentist is increased due to the small operating field, frequent patient movements, and the variety of sharp instruments used in dental procedures - burs, scalers, scalpels and endodontic files [1]. Hepatitis viruses (A, B and C) can be detected in oral fluids including whole saliva and gingival crevicular fluid [2]. Patients with chronic HCV infection exhibit more severe periodontitis, including gingival bleeding, increased pocket depth and attachment loss, when compared to healthy controls [3]. Studies on the epidemiology of HCV have demonstrated its low infectivity in saliva [4]. Also, in drops generated during dental procedures, HCV can survive up to 6 weeks, but in low titers [5].

HBV infection can present with extrahepatic manifestations including the oral cavity. The infection can affect the salivary glands, leading to xerostomia and sialadenitis [6]. The gingival sulcus has been identified as the intraoral location with the greatest concentration of HBV [7]. Hepatitis B surface antigen (HBsAg) was detected in the gingival crevicular fluid and saliva samples of 90% of HBV infected patients [8]. It was demonstrated that HBV DNA detection in saliva samples is detected more often in patients with detectable HBsAg and hepatitis B e-antigen (HBeAg) in serum [9]. Also, HBV is characterized with prolonged environmental stability, which was confirmed by animal infection model where positive for HBV human

plasma dried for 1 week and inoculated in chimpanzees resulted in an active infection [10].

Despite the paucity of data, Bulgaria is characterised by a low prevalence of hepatitis C, with an overall anti-HCV prevalence of 1.3%, ranging from 0.7% to 1.6% [11]. The country also exhibits intermediate prevalence of hepatitis B, with a crude rate of 3.9% for HBsAg [12]. According to the latest epidemiological data from the National Center for Infectious and Parasitic Diseases (NCIPD), the incidence of acute viral hepatitis B in 2024 was 4.06 per 100 000, while the incidence of viral hepatitis C was found 1.78 per 100 000. The relative share of the two infections was determined to be 0.49 and 0.22, respectively [13; access date: 27/08/2025]. The screening of healthcare workers (HCWs) for hepatitis B is carried out following Regulation No. 15 from May 2005 on the implementation of vaccination in the Republic of Bulgaria. According to the stipulations of the Terms and Conditions for Recommended Immunizations, it is strongly recommended that medical and non-medical professionals, including service personnel operating within medical and healthcare facilities, undergo immunization against viral hepatitis B through the administration of a recombinant hepatitis B vaccine. This recommendation is particularly applicable to medical and dental students enrolled in higher medical educational institutions, provided they are negative for HBsAg and do not possess any laboratory-confirmed data substantiating naturally acquired or post-vaccination immunity to HBV. This recommendation is further reinforced by Regulation No. 4 of 2002, which stipulates the protection of workers from risks associated with exposure to biological agents at the workplace.

Therefore, dental professionals need special consideration for hepatitis B and hepatitis C screening, and HBV vaccination. Despite the implementation of effective HBV vaccination programs, which have reduced the risk of HBV transmission among HCWs, particularly among DHPs, the evaluation of hepatitis B surface antibody (HBsAb) post-vaccination response remains crucial. The objective of the present study was to estimate the prevalence of serological markers for hepatitis B and hepatitis C among practicing dental healthcare professionals.

MATERIALS AND METHODS

2.1. Design

A cross-sectional survey was conducted to evaluate the presence of serological markers for hepatitis B and C among dental healthcare professionals. The study was conducted between 1 June and 31 October 2024 as a part of a larger sero-epidemiological study on the prevalence of hepatitis B and hepatitis C serological markers among HCWs aunder the National Program for Prevention and Control of Viral Hepatitis in the Republic of Bulgaria 2021–2025. Prior to participation, all subjects provided a written informed consent. The survey protocol was approved by the Expert Advisory Council on Viral Hepatitis at the Bulgarian Ministry of Health (MH).

2.2. Sampling

The calculation of the quota samples for the 28 administrative regions in Bulgaria was based on statistical data for registered HCWs, DHPs included, as published by the National Statistical Institute (NSI) of Bulgaria for 2023 [14]. Due to the significantly smaller number of registered DHPs, they were grouped together with HCWs, and the quota sample for each Regional Health Inspectorate (RHI) was determined

Table 1. Distribution of recruited DHPs participants by RHIs

Regional Health Inspectorate	Calculated quota based on the total number of HCWs	Actual number of recruited DHPs
Blagoevgrad	70	0
Burgas	80	0
Varna	110	29
Veliko Tarnovo	70	10
Vidin	60	7
Vratsa	60	5
Gabrovo	60	10
Dobrich	60	0
Kardzhali	60	10
Kyustendil	60	4
Lovech	60	1
Montana	60	0
Pazardzhik	60	0
Pernik	60	0
Pleven	80	0
Plovdiv	180	11
Razgrad	60	1
Ruse	60	0
Silistra	60	0
Sliven	60	10
Smolyan	60	10
Sofia Province	60	0
Sofia Capital	215	0
Stara Zagora	80	0
Targovishte	50	4
Haskovo	60	18
Shumen	60	3
Yambol	50	0

based on the total number. At the commencement of the study, each RHI was provided with all the requisite documentation, including an official letter from the MH of the Republic of Bulgaria for conducting a seroepidemiological study, a list of quota samples, an informed consent form, a questionnaire for participants, and instructions for sample collection and transport. The decision regarding the collection of samples was delegated to the RHIs. Following the distribution of an invitation letter, the following options for the collection of samples were made available: 1) at the RHI site, or 2) at the hospital/medical centre site. During the sampling, participants self-completed a questionnaire including: socio-demographic data, professional background, history of hepatitis testing, and HBV vaccination status. The voluntary nature of the study resulted in a final number of 133 collected samples . The distribution of participants by RHIs is presented in Table 1.

2.3. Detection of serological markers for hepatitis B and hepatitis C

A qualitative enzyme immunossay was used to determine the presence of antibodies against HCV (HCV Ab v.4 ELISA; DIAPRO, Italy), and HBV surface antigen (HBsAg one v. Ultra ELISA; DIAPRO, Italy). For both tests, samples with a serological index (the ratio between the sample's optical density at a wavelength of 450 nm and the Cutt-Off value) of ≥ 1.1 were designated as positive. A qualitative chemiluminescent microparticle immunoassay (CMIA) was used for the detection of antibodies against hepatitis B core antigen (INNODX HBcAb CMIA; Xiamen Innodx Biotech Co, P.R. of China). The analysis was based on the sandwich principle to detect antibodies to HBV core antigen in human serum and plasma. All samples with the calculated index > 1.0 were considered positive. For the detection of antibodies against HBsAg a quantitative enzyme immunoassay was performed (HBsAb; DIAPRO, Italy). The calculation of HBsAb concentration was performed using a calibration curve derived from five calibrators at 0 IU/ml, 10 mIU/ml, 50 mIU/ml, 100 mIU/ml and 250 mIU/ml. HBsAb titer of >10 mIU/ml was considered positive.

2.4. Statistical analysis

Continuous data were expressed as median with interquartile range (IQR – 25^{th}; 75^{th}). Numbers and percentages (n, %) were used to present qualitative variables. To evaluate disparities between proportions, a z-test was conducted, comparing the predominant proportion with the remaining proportions within the corresponding group. A 2-sided p-value of <0.05 was considered statistically significant. Statistical analyses were performed using the online Epitools Epidemiological Calculators – Ausvet [15] and SPSS Statistics for Windows, v.25 (SPSS Inc., Chicago, Ill., USA).

RESULTS

3.1. Socio-demographic characteristics of the study population.

The study population of DHPs included 133 participants, from whom 55% were dentists, 14% were dental assistants and nurses (p=0.0018), 8% were dental technicians, laboratory technicians, and sanitarians (p = 0.0036), and 23% were administrators and participants

Table 2. Main characteristics of the DHPs population

	N	%	*z-value*	*p-value*
occupation (N=133)*				
dentists	73	55		
dental assistants, nurses	18	14	3.1	.0018
dental technicians, lab.technicians, sanitarians	11	8	2.9	.0036
administrators, others	31	23	3.0	.0027
age [decades] (N=133)				
<30	29	22	0.9	
30-39	29	22	0.9	
40-49	28	21	0.9	NS
50-59	25	19	0.8	
60-69	17	13	0.6	
≥70	5	4		
sex (N=133)				
male	23	17	6.4	< .0001
female	110	83		
year of entry into the healthcare system (N=104)*				
<=1990	20	19	0.3	
1991-2000	22	21	0.2	
2001-2010	22	21	0.2	NS
2011-2020	24	23		
>2020	16	15	0.6	
HBsAg and HCV Ab presence (N=133)				
HBsAg (+)	0	0	--	NA
HCV Ab (+)	0	0	--	

Legend: *Denominator totals vary due to incomplete responses; NS = not significant; A comparison was made between the predominant proportion and the remaining proportions within each respective analyzed feature. Significance level 0.05

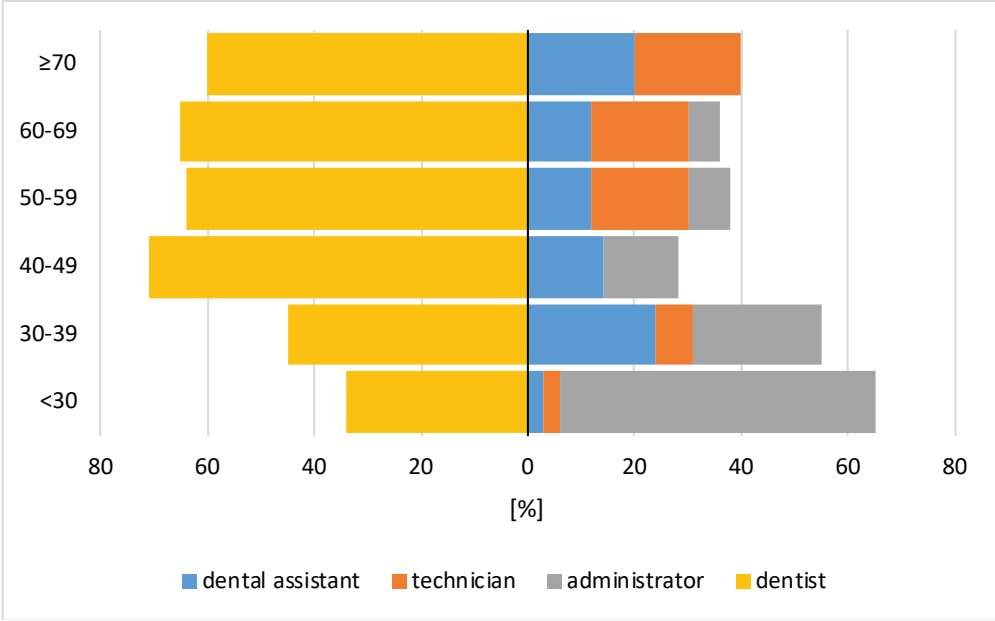

Figure 1. Age distribution of DHPs according their occupation

who self-reported as others (p=0.0027) (Table 2). The age of the enrolled DHPs was (median, IQR) 43 (30.5; 54) years. ranging from 20 to 73 The demographic composition of the study population was almost equally represented across all age groups (ranging from younger than 30 years to 60 - 69 age group), with the exception of the oldest participants (aged ≥70), who constituted 4% of the total population. However, for the dentist the highest percent of participants was in age group 40-49 years (71%), for dental assistants – in age group 30-39 (24%), for administrators - in age group <30 (59%), and for technicians the most represented age group was ≥70 years –with 20% (Figure 1).

Women outnumber men almost fivefold , 83% vs.17%, respectively (p < .0001). The longest presence in the healthcare system was recorded since 1988, and the shortest - since 2024. None of the participants tested positive for HBsAg or HCV Ab.

3.2. Professional accidents among dental healthcare professionals.

Of 133 enrolled DHPs 132 answered the question about professional accident (Table 3). The confirmatory answers were 17% (p <.0001). Dentists were the most affected, with a statistically significant difference ascompared to the group of dental assistants and nurses (74% vs 17%, p=0.0335). From 23 DHPs, who reported a professional accident, 78% (p=.0195) were vaccinated against HBV. On the other hand, 52% of DHPs, who self-reported a history professional accident, denied having undergone a previous hepatitis testing. At the same time, the duration of profession-

Table 3. Correlation between professional accidents and occupied position, HBV vaccination status, previous hepatitis testing and profesional experience duration

	N	%	*z-value*	*p-value*
professional accident, total (N=132)				
Yes	23	17	6.4	< .0001
No	109	83		
occupation (N=23)				
dentist	17	74		
dental assistant, nurse	4	17	2.1	.0335
dental technician, lab technician, sanitarian	1	4	--	NA
administrator, others	1	4	--	
HBV vaccination status among DHPs who suffered professional accident (N=23)				
Yes	18	78	2.3	.0195
No	5	22		
previous hepatitis testing in DHPs self-reported professional accident (N=23)				
Yes	11	48	0.2	NS
No	12	52		
year of entry into the healthcare system (N=23)				
<=1990	5	22	--	NA
1991-2000	5	22	--	
2001-2010	5	22	--	
2011-2020	5	22	--	
>2020	3	13	0.3	NS

Legend: DHPs=dental healthcare professionals; NA = not applicable; NS = not significant. A comparison was made between the predominant proportion and the remaining proportions within each respective analyzed feature. Significance level 0.05

al experience exhibited no correlation with the occurrence of professional accidents.

3.3. HBV vaccinated status in dental healthcare professionals

A total of 132 DHPs provided information regarding their HBV vaccination status (Table 4). The proportion of vaccinated participants was not significantly different from the proportion of non-vaccinated participants (49% vs. 51%, respectively). The most recent vaccination was reported one year prior by 5% of the participants. The majority of respondents (79%) reported receiving the vaccination more than a decade ago. The presence of protective HBsAb was detected in 49% of the DHPs who self-reported as vaccinated, as the time when they were vaccinated against hepatitis B was predominantly more than two years ago. The presence of HBsAb was detected in 25% of the non-vaccinated participants.

Table 4. Vaccination status of the DHPs population

	N	%	*z-value*	*p-value*
HBV vaccine, total (N=132)				
Yes	65	49	0.2	NS
No	67	51		
Time of vaccination (N=63)*				
The current year	0	0	--	--
1 year ago	3	5	2.9	.0041
2 years ago	0	0	--	--
> 2 years ago	4	6	3.2	.0014
> 5 years ago	6	10	3.5	.0004
> 10 years ago	50	79		
detection of HBsAb in vaccinated DHPs (N=65)				
HBsAb (+)	32	49	0.2	NS
HBsAb (-)	33	51		
time of vaccination of HBsAb positive DHPs (N=32)				
the current year	0	0	--	--
1 year ago	2	6		
before 2 years	0	0	--	--
> 2 years	3	9	2.3	.0212
> 5 years	3	9		
> 10 years	24	75		
detection of HBsAb in non-vaccinated DHPs (N=67)				
HBsAb (+)	17	25	3.7	.0002
HBsAb (-)	50	75		

Legend: *Denominator totals vary due to incomplete responses; DHPs=dental healthcare professionals; NS = not significant. A comparison was made between the predominant proportion and the remaining proportions within each respective analyzed feature. Significance level 0.05

3.4. Measured HBsAb and HBcAb status of dental healthcare professionals

The actual HBsAb status of 133 DHPs was determined (Table 5). For 37% of the participants, positive results were established (z=2.9, p=.0037). In 49% of the cases, the titer of HBsAb was found to be greater than 100 mIU/ml. In a subset of 10% of all HBsAb-positive DHPs, the presence of HBcAb was detected (z=4.4, p<.0001). Of these subjects, 20% reported being vaccinated against HBV. Dentists demonstrated the highest prevalence of past HBV infection, with 80% of the samples exhibiting positive results.

Table 5. HBsAb and HBcAb status of dental healthcare professionals

	N	%	*z-value*	*p-value*
detection of HBsAb, total (N=133)				
HBsAb (+)	49	37	2.9	.0037
HBsAb (-)	84	63		
HBsAb titer [mIU/ml] (N=49)				
< 100	25	51	0.1	NS
> 100	24	49		
detection of HBcAb in HBsAb positive DHPs (N=49)				
HBsAb(+)/HBcAb(+)	5	10	4.4	<.0001
HBsAb(+)/HBcAb(-)	44	90		
vaccination status of the HBsAb(+)/HBcAb(+) DHPs (N=5)				
vaccinated	1	20	--	NA
not vaccinated	4	80		
occupation by the HBsAb(+)/HBcAb(+) DHPs (N=5)				
dentist	4	80	--	NA
dental assistant, nurse	0	0		
dental technician, lab technician, sanitarian	1	20		
administrator, others	0	0		

Legend: DHPs=dental healthcare professionals; NA = not applicable; NS = not significant. A comparison was made between the predominant proportion and the remaining proportions within each respective analyzed feature. Significance level 0.05

Discussion

The recent study complements other publications on hepatitis B and C in the context of dentistry in Bulgaria. While others have focused on the oral health of patients with chronic viral hepatitis [7, 16], or on the knowledge regarding viral hepatitis [17], a recent analysis assessed serological markers for viral hepatitis B and C among enrolled practicing dentists. The

present sero-epidemiological study clearly demonstrates an uneven distribution of dental healthcare professionals by gender, with women outnumbering men by almost fivefold. A general absence of statistically significant differences between the age groups is indicated. However, a predominance of dental practitioners in the age groups over 40 years was observed. According to the survey results, 17% of the participants reported having experienced a professional accident, with dentists exhibiting a higher prevalence of such incidents. With respect to the serological markers of hepatitis, the presence of antibodies against HCV was not detected. In the survey, 49% of the DHPs reported a vaccination for hepatitis B. Among those who received the vaccination, approximately 50% exhibited protective antibodies, as over 70% reported receiving vaccination within the past decade. The serological patterns of past HBV infection were identified in 10% of the study participants. Notably, only 20% of these DHPs reported a vaccination against HBV.

This study was the result of collaborative efforts by various participants, including the Ministry of Health, the NRL of Hepatitis viruses and 28th RHIs from across the country. As a result, the sampling stage was subject to variation. Despite the efforts made by each RHIs, the number of participants who voluntarily enrolled in the study was considerably low, with a total of 133 DHPs (who reported their occupation in the dental field and/or place of work in a dental centre). A key challenge in conducting seroepidemiological studies is the inadequate number of participants. For instance, in a pilot study on the prevalence of HCV among the general population, active participation reached 21.6% [11]. Also, the participation in surveys was found to be dependent not only on itheir design but on the sociodemographic characteristics of the participants, including age, sex, country of origin, education, and labor market attachment [18]. In the present cross-sectional survey, dentists constituted the most prevalent professional category among the enrolled participants. The oldest age group (>70 years) was the least represented, which can be attributed to the reduced time presence of this age group at the workplace.

Due to the nature of their work, DHPs are subject to an elevated risk of sharp injuries. It was established that despitethe implementation of safety prevention measures, sharp injuries related to dental explorers and dental injection needles remain a common type of injury [19]. A meta-analysis of global data revealed the prevalence of needle-stick injuries among dentists to be between 27.5% and 69.2%, with a pooled prevalence of 59.1%, which was the highest among healthcare workers based on their job type [20]. Similarly, recent cross-sectional survey has revealed that the prevalence of self-reported professional accidents was the highest among dentists (74%), followed by dental technicians and nurses. Notably, approximately half of affected DHPs reported not having undergone prior hepatitis testing.

Both HBV and HCV can be detected in saliva, despite the fact that oral transmission of HCV has not been confirmed [21]. This finding may provide a rationale for the negative results observed for HCV antibodies in all the tested samples from the resent survey. The transmission of HBV through saliva has been confirmed in an animal infectious model [22]. Furthermore, the presence of HBV DNA is detected in 80% of patients with occult HBV infection [23], thereby underscoring the potential risk for dentists to contract HBV. This correlation was also confirmed in the present study, as dentists had the highest percentage of samples positive for HBcAb, which is a marker for past HBV infection.

The most effective strategy to prevent HBV infection is vaccination. The protection provided by three or four doses of hepatitis B vaccine can persist for a period of at least two decades [24] and the HBsAb levels ≥ 10 mIU/ml are considered protective [25]. The present study revealed that 49% of the DHPs reported receiving a vaccine for hepatitis B. However, only half of these participants had detectable HBsAb. At the same time, over 70% of subjects reported having received the vaccination for a period more than 10 years prior. A study of healthcare workers has established that the cumulative persistence of HBsAb 18 years after vaccination was 76.5% for high responders (1 month after complete vaccination HBsAb > 1000 mIU/ml), 35.4% for medium responders (100-999 mIU/ml) and 23.5% for low responders (10-99 mIU/ml) [26]. This can explain the decline in HBsAb to undetectable levels. At the same time, the present study identified that 25% of DHPs, who reported not having been vaccinated had protective antibodies. These antibodies can be attributed to the inaccurate assessment of vaccination status by the participants. According to the European recommendations for the

management of healthcare workers occupationally exposed to the hepatitis B virus, the presence of post-vaccinal HBsAb levels of at least 10 mIU/ml is indicative of responders [25]. More recent publications have demonstrated that an HBsAb titer above 100 IU/L is indicative of protective immunity [27]. In the recent study, only half of the enrolled DHPs had an antibody titer ≥ 100 mIU/ml. At the same time, in the cohort of DHPs for which HBsAb had been detected, 10% were found to be cumulatively positive for HBcAb. The results suggest a history of HBV infection. Comparable percentages of HBcAb positivity have been documented among dental healthcare workers and by other authors, as the value range from 4.97% [28] to 12.1% [29]. As demonstrated in the study conducted in Japan, the positive rate of HBcAb exhibited an age-related increase, from 2.9% among individuals aged 30-39 to 5.6% among those aged 40-49, 29.4% among those aged 50-59, and 85.7% among those aged 60-69, as 25% was not vaccinated [29].

In summary, to our knowledge, this is the first study conducted to provide a comprehensive overview of the prevalence of serological markers for hepatitis B and C among practicing dental healthcare professionals. Despite the limited number of participants enrolled in the study, the approach adopted was efficacious in estimating the prevalence of serological markersf for hepatitis B and C among practicing DHPs. The findings of this study suggest an enhanced screening for viral hepatitis among dental professionals as well as the development of technical guidelines for the prevention of hepatitis B and C infection among dental professionals.

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Declaration of interests

The authors declare no Conflicts of interest.

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