

Immunization against Hepatitis B Virus: Serological Status of West African Populations Residing in Cotonou (Benin) and Dakar (Senegal)

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Abstract

Background: According to WHO estimates, by 2022 over 296 million people are living with chronic hepatitis B virus (HBV) infection, and over 820,000 have died from complications. In sub-Saharan African countries such as Benin and Senegal, few research studies have addressed the issue of HBV immunization. **Objective:** The main objective of this study was to evaluate immunization against the hepatitis B virus in populations residing in Cotonou and Dakar by titrating anti-HBs antibodies (Ab) and detecting total anti-HBc immunoglobulins (Ig). **Materials and Methods:** This was a prospective, descriptive, analytical study of two West African populations recruited in Dakar at the Laboratory of Medical Biology (LBM) of the General Hospital Idrissa Pouye (HOGIP) and in Cotonou at the LBMs of the health centres of the Cotonou archdiocese. HBsAg-negative patients constituted our study population. The study took place in November-December 2019 for Dakar and February-March 2020 for Cotonou. Anti-HBs antibodies were tested and titrated. In the event of anti-HBs positivity, total anti-HBc was determined. A microparticle chemiluminescence immunoassay was used for marker determination. The detection threshold was 2.50 IU/L for anti-HBs. Excel and IBM SPSS Statistics software were used for data analysis. Subjects' sociodemographic characteristics were collected using a questionnaire, as was knowledge of their vaccination status. The study was approved by the ethics committees in Benin and Senegal. **Results:** A total of 394 HBs antigen-negative

participants were recruited: 205 in Cotonou and 189 in Dakar. The population was predominantly female, with 65.36% (N = 134) and 57.14% (N = 108) women in Cotonou and Dakar respectively. The median age of participants was 29 years in Cotonou, with extremes of 10 and 65 years, versus 39 years in Dakar, with extremes of 6 and 93 years. Some participants claimed to be unaware of their vaccination status: 33.17% in Cotonou and 56.61% in Dakar. The total prevalence of anti-HBs-positive subjects was 88.78% (N = 182) in Cotonou and 98.41% (N = 186) in Dakar. In Cotonou (N = 205), 35.61% (N = 73) of subjects had protective anti-HBs levels between 11.60 IU/L and 10,000 IU/L. In Dakar, 61.38% (N = 116) of subjects had protective HBV immunity, with anti-HBs titres ranging from 10.30 IU/L to 11357 IU/L. In Cotonou, 80.82% (N = 59) of immunized subjects (N = 73) had anti-HBc antibodies, compared with 84.48% (N = 98) of immunized individuals (N = 116) in the population recruited in Dakar, indicating immunization following HBV infection. **Conclusion:** Our study involved a predominantly female population, many of whom were unaware of their serological status. Vaccination policies and knowledge of the viral hepatitis B epidemic need to be strengthened.

Keywords

HBV, Anti-HBs, Anti-HBc-Cotonou-Dakar

1. Introduction

Hepatitis B is an inflammation of the liver caused by the hepatitis B virus (HBV). HBV infection is usually asymptomatic, and goes undetected in adults in 95% of cases. In children, on the other hand, the condition is chronic and progresses to cirrhosis and primary liver cancer [1]. HBV is a major public health problem, particularly in sub-Saharan Africa.

In 2022, epidemiological data from the World Health Organization (WHO) estimated the number of people living with chronic HBV infection at 296 million, with over 820,000 deaths due to complications of the disease [2]. Most people living with Hepatitis B Virus (HBV) are unaware of their immune status and live in endemic regions [3].

Many research studies have addressed several aspects of viral hepatitis B, but few have focused specifically on the issue of immunization against the virus.

Senegal and Benin are among those countries where the epidemiological data available are not very varied, and mostly concern seroprevalence. Compulsory vaccination at birth is a very recent development in West Africa. A few previous studies have reported low HBV seroprotection rates in the West African sub-region [4] [5]. It is in this context that our study focuses on immunization against hepatitis B virus in the populations of the countries concerned. This study will thus contribute to completing and diversifying the available data on viral hepatitis B in Benin and Senegal, with the aim of better understanding the problem of the level of protection of the populations against HBV. The main objective of

this study was therefore to assess immunization of populations against the hepatitis B virus in Cotonou and Dakar by titration of anti-HBs and detection of total anti-HBc.

2. Materials and Methods

2.1. Study Population

This was a prospective, descriptive study of Hepatitis B Virus (HBV) immunization in a cohort of participants recruited at the Medical Biology Laboratory (LBM) of the General Hospital Idrissa Pouye (HOGIP) in Dakar and at the LBMs of the health centres of the Archdiocese of Cotonou.

Our study population consisted of HBs-antigen-negative individuals derived from a previous study of hepatitis B seroprevalence in the same populations. Initial recruitment was carried out in three phases: first, the reception of all the patients met in the medical biology laboratories for a general presentation of the object of the research; then the individual reception of the interested patients for a detailed presentation of the steps of the research in order to obtain their consent; finally, the collection of the socio-demographic data of each patient by means of a questionnaire and the collection of venous blood on a dry tube.: The size of the initial sample was estimated using Schwartz's formula.

It took place in November-December 2019 for Dakar and February-March 2020 for Cotonou. All biological tests were performed at LBM-HOGIP in Dakar. Our study population consisted of all consenting HBs antigen-negative individuals encountered at the recruitment sites during the different periods. The initial criterion for subject selection was presence at the recruitment sites for a blood test.

The following were not included in this study:

- Subjects already undergoing treatment for chronic HBV infection;
- Subjects who refused to participate in the study.

2.2. Ethical Considerations

The present study received ethical approval from the Research Ethics Committee (CER) of the Cheikh Anta Diop University (UCAD) of Dakar under number 041012019/CER/UCAD and from the National Ethics Committee for Health Research (CNERS) of Benin under number 48/November 2019. Throughout the study, the commitments made to the various ethics committees were respected from data collection to publication of the results. In accordance with the recommendations of the ethics committees, we required the assent of minors, as well as the consent of their parent or guardian.

2.3. Data Collection

Data were collected on socio-demographic characteristics (first name, surname, sex, age, marital status, occupation, level of education) and on the population's knowledge of their hepatitis B vaccination status. A database for each study

population was created using Excel Office 365.

2.4. Analytical Method

Serological tests were performed using microparticle chemiluminescence immunoassay technology for the titration of anti-HBs and the qualitative detection of total anti-HBc. Total anti-HBc antibodies were tested in anti-HBs-positive subjects. The detection threshold for the anti-HBs assay is 2.50 IU/L. Chemiluminescence microparticle technology is adapted to an integrated automated platform for clinical chemistry and immunology testing (Architect Ci4100 from Abbott Diagnostics, USA). Calibrations were carried out, validated and stored in the system prior to analysis of internal quality controls and test samples. Typical values for anti-HBs titres in people who have been in contact with HBV range from 2.50 IU/L to 1000 IU/L.

2.5. Statistical Analysis of Data

Statistical analysis of the data was performed using Excel Office 365 and IBM SPSS Statistics version 20. The statistics used to describe and analyse the variables were percentage, mean, standard deviation and median. Chi-square tests, analyses of variance, unpaired Student's t-tests or non-parametric tests (Mann-Whitney, Spearman) were used to compare patient characteristics. In all analyses, a threshold value of $p < 0.05$ was considered statistically significant.

3. Results

3.1. Characteristics of the Study Population

Our study included 394 participants with negative HBs antigenemia: 205 in Cotonou, 65.4% of whom were women ($N = 134$), and 189 in Dakar, 57.1% of whom were women ($N = 108$). The median age was 29 years in Cotonou, with extremes of 10 and 65 years, versus 39 years in Dakar, with extremes of 6 and 93 years (**Table 1**).

3.2. Vaccination Status of Study Populations

During the recruitment survey, we asked participants about their knowledge of their hepatitis B vaccination status. In Cotonou ($N = 205$), 25 participants thought they had been vaccinated, compared with 180 who thought they had never been vaccinated (**Table 2**). In Dakar ($N = 189$), 21 participants claimed to have been vaccinated versus 168 who said they had never been vaccinated (**Table 3**).

Declarations of vaccination status contradicted the results of anti-HBs titration. In Cotonou ($N = 205$), 32.22% ($N = 58$) of participants who thought they had never been vaccinated ($N = 180$) had anti-HBs titres above 10 IU/L, while 40% ($N = 10$) of those who thought they had been vaccinated ($N = 25$) had anti-HBs titres below 10 IU/L, (**Table 2**). In the population recruited in Cotonou ($N = 205$), 33.17% ($N = 68$) were unaware of their vaccination status. In the

Table 1. Main characteristics of the population.

Variables	Population recruited in Cotonou					Population recruited in Dakar				
	Men		Women		Total	Men		Women		Total
	N	% (CI) 95%	N	% (CI) 95%		N	% (CI) 95%	N	% (CI) 95%	
Total	89	38 [0.31; 0.45]	145	62 [0.55; 0.69]	234	112	47.46 [0.40; 0.54]	124	52.54 [0.45; 0.59]	236
Seroprotection markers N = 394										
anti HBs Ab	71	34.6 [0.28; 0.41]	134	65.4 [0.58; 0.72]	205	81	42.9 [0.36; 0.50]	108	57.1 [0.50; 0.64]	189
Seroprotected individuals: Anti HBs Ab ≥ 10 IU/L	32	43.8 [0.37; 0.51]	41	56.2 [0.49; 0.63]	73	48	41.4 [0.34; 0.48]	68	58.6 [0.51; 0.66]	116
Non seroprotected individuals: AntiHBs Ab < 10 IU/L (non zero)	30	27.5 [0.21; 0.35]	79	72.5 [0.66; 0.80]	109	33	47.1 [0.41; 0.55]	37	52.9 [0.46; 0.60]	70
Non infected and unvaccinated individuals: Ag HBs and anti HBs Ab negative	9	39.13 [0.26; 0.40]	14	60.87 [0.60; 0.74]	23	0	0 [-0.07; 0.07]	3	100 [0.93; 1.07]	3

Legend: N = Number, CI = Confidence Interval; % = Percentage.

Table 2. Vaccination status of the population recruited in Cotonou.

	Vaccination statut	Anti-HBs Ab < 10 IU/L	Anti HBs Ab > 10 IU/L
Unvaccinated	180	67.78% (N = 122)	32.22% (N = 58)
Vaccinated	25	40% (N = 10)	60% (N = 15)
Total	205	132	73

Legend: **Unvaccinated** = participant who said he/she had never been vaccinated against HBV; **Vaccinated** = participant who said he/she had received at least one dose of hepatitis B vaccine; % = percentage; N = number.

Table 3. Vaccination status of the population recruited in Dakar.

	Vaccination status	Anti-HBs Ab < 10 IU/L	Anti-HBs Ab > 10 IU/L
Unvaccinated	168	39.88% (N = 67)	60.12% (N = 101)
Vaccinated	21	28.57% (N = 6)	71.43% (N = 15)
Total	189	73	116

Legend: **Unvaccinated** = participant who said he/she had never been vaccinated against HBV; **Vaccinated** = participant who said he/she had received at least one dose of hepatitis B vaccine; % = percentage; N = number.

population recruited in Dakar (N = 189), 60.12% (N = 101) of participants who said they had never been vaccinated against HBV (N = 168) had anti-HBs titres above 10 IU/L, while 28.57% (N = 6) of those who said they had been vaccinated (N = 21) had anti-HBs titres below 10 IU/L (Table III). In the population recruited in Dakar, 56.61% (N = 107) were unaware of their vaccination status.

3.3. Immunization against Hepatitis B Virus

With regard to HBV immunization, out of a total of 394 participants, 26 were anti-HBs negative (zero anti-HBs titre). Considering anti-HBs negative subjects, 23, 39.13% of whom were men (N = 9), were from the population residing in Cotonou, and 3 others, all female, were from the population recruited in Dakar (**Table 1**).

In the Cotonou study (N = 205), 35.61% (N = 73) of subjects had protective immunity against HBV, with anti-HBs titres between 11.60 IU/L and 10000 IU/L, versus 64.39% (N = 132) who had non-protective anti-HBs titres, *i.e.* below 10 IU/L and zero (**Table 4**). Female subjects accounted for 56.2% (N = 41) of those protected against HBV (N = 73) (**Table 1**). In this population recruited in Cotonou, a majority of 80.82% (N = 59) of subjects with anti-HBs titres above 10 IU/L (N = 73) carried total anti-HBc, while 19.18% (N = 14) did not express anti-HBc (**Table 5**). The median anti-HBs titre in HBV-protected subjects (N = 73) was 139 IU/L, (**Table 5**).

In the Dakar study (N = 189), 61.38% (N = 116) of subjects had protective immunity against HBV, with anti-HBs titres between 10.30 IU/L and 11537 IU/L, versus 38.62% (N = 73) who had anti-HBs titres below 10 IU/L and zero (**Table 6**). Female subjects accounted for 58.6% (N = 68) of those protected against HBV (N = 116) (**Table 1**). A majority of 84.48% (N = 98) of subjects with anti-HBs titres above 10 IU/L (N = 116) were total anti-HBc positive, and 15.52% (N = 18) were total anti-HBc negative (**Table 7**). The median anti-HBs level in protected subjects (N = 116) was 98.65 IU/L, (**Table 6**).

Table 4. Anti-HBs titration for the population living in Cotonou.

	N	%
Anti HBs Ab < 10 IU/L (Non protected)	132	64.39
Anti HBs Ab ≥ 10 IU/L (Immunized)	73	35.61
Total	205	100

Median immunized patients: 139 IU/L; Legend: N = number; % = percentage; 64.39% of the population had anti-HBs titres below 10 IU/L and 35.61% had anti-HBs titres greater than 10 IU/L.

Table 5. Anti-HBc and anti-HBs titration for the protected population living in Cotonou.

	N	%
Anti HBs Ab ≥ 10 UI/L & Anti-HBc Ab negative	14	19.18
Anti HBs Ab ≥ 10 UI/L & Anti-HBc Ab positive	59	80.82
Total	73	100

Legend: N = number; % = percentage; 19.18% had anti-HBs titres greater than 10 IU/L and also carried anti-HBc, while 80.82% had anti-HBs titres greater than 10 IU/L and did not carry anti-HBc.

Table 6. Anti-HBs titration for the population recruited in Dakar.

	N	%
Anti HBs Ab < 10 UI/L (Non protected)	73	38.62
Anti HBs Ab > 10 UI/L (Immunized)	116	61.38
Total	189	100

Median patients immunized: 98.65 UI/L; **Legend:** N = number; % = percentage; 38.62% had anti-HBs titres below 10 IU/L and 61.38% had anti-HBs titres above 10 IU/L.

Table 7. Anti-HBc and anti-HBs titration, protected population living in Dakar.

	N	%
Anti-HBs Ab \geq 10 IU/L & Anti-HBc Ab negative	18	15.52
Anti-HBs Ab \geq 10 IU/L & Anti-HBc Ab positive	98	84.48
Total	116	100

Legend: N = number; % = percentage; 15.52% had anti-HBs titres above 10 IU/L and did not carry anti-HBc, while 84.48% had anti-HBc titres above 10 IU/L and carried anti-HBc.

4. Discussion

We opted for a prospective, descriptive, analytic study with a questionnaire, as this approach favors meeting with the subjects in order to obtain their individual consent, as required by the ethics committees in Benin and Senegal. In addition, a personalized meeting with the participants was necessary to collect certain information relating to socio-demographic data and their vaccination status.

In our study, the chemiluminescence analytical technique was chosen in preference to other methods because of its efficacy according to the work of Laouina *et al.* in 2016 [6].

Our study revealed a female majority in the population in both Cotonou and Dakar. Generally speaking, women make up the majority in the demand for healthcare. The female majority in hospital studies has indeed been reported in some research works such as those conducted by Ahmed Ben *et al.* on the demand for care in general medicine in Tunisia in 2004 [7]. In another study conducted by Soliman *et al.* in 2016 on mass screening for hepatitis B and C in southern Egypt, women made up 52.3% of the population [8].

In our populations recruited in both Cotonou and Dakar, many participants were unaware of their vaccination status. In fact, in our populations, some doubted their hepatitis B vaccination status, while a larger number were almost completely unaware of viral hepatitis B. This lack of knowledge of viral hepatitis B in sub-Saharan African populations has indeed been reported in previous studies, including those carried out by Lawson-Ananissoh L. *et al.* in 2015 on patients consulting gastroenterology in Lome (Togo). The same observation was made in research carried out in western Ghana in 2020 by Anfaara N. *et al.* [9] [10]. In another study carried out by Okonkwo U. *et al.* in Nigeria in 2018, over

50% of the population were unaware of viral hepatitis B, its pathogen, transmission routes, even symptoms, etc. [11]. We can deduce from this that viral hepatitis B remains very little known in the populations of sub-Saharan Africa, particularly in unschooled areas.

In terms of HBV protection, only 35.61% of subjects recruited in Cotonou had protective anti-HBs titres (anti-HBs > 10 IU/L). In this Cotonou population, 80.82% of subjects were protected after a resolved HBV infection, as revealed by the presence of total anti-HBc in their serum. In the Dakar study, 61.38% of subjects were protected against HBV. In this Dakar resident population, 84.48% of immunized subjects were protected after a resolved infection, as they are total anti-HBc positive. The low rates of protection against HBV in our study reflect the inadequacy of vaccination coverage and ignorance of the practice of means of protection among our populations. These rates of protection found in our study are in harmony with data from some previous work on different populations. Indeed, the HBV seroprotection rate according to research conducted by Denis F. *et al.* on Beninese soldiers sent on mission to Ivory Coast in 2019 was 23.42% [12]. Research conducted in 2017 by Niang M. *et al.* on pregnant women in Dakar suggested a seroprotection rate of 40% [13].

The rates of protective immunization against HBV in our various study populations, being higher than those reported in previous studies of similar general populations, would nevertheless reflect progress in vaccination coverage. Indeed, a study by Lô G. *et al.* of children in the Dakar region of Senegal in 2019 reported a seroprotection rate of 76.6% [14]. The introduction of the HBV vaccine into the Expanded Program on Immunization is actually recent in both countries: 2002 for Benin and 2005 for Senegal, starting six weeks after birth [4] [14].

Finally, the fact that most of the participants protected against HBV were protected after a resolved HBV infection in both Cotonou and Dakar draws attention to the necessity and urgency of vaccination within twenty-four hours of birth, as recommended by the WHO to break the chain of transmission at the base [3]. The rate of seroprotection against HBV is therefore increasing in our populations. With 24-hours vaccination only becoming effective in 2016 in Senegal and later in 2020 in Benin, we can hope that the impact of these vaccination programs will be increasingly perceptible in the years to come, with rising seroprotection rates.

There are a number of limitations to this study as a whole. Firstly, as participants undergoing vaccination were not followed up over a period of time, this study did not make it possible to assess anti-HBs antibody synthesis over time after each vaccine dose. This would have allowed us to assess the relevance of booster doses of HBV vaccine. Secondly, this study would have been better able to assess progress in vaccination coverage in the different countries if it had specifically targeted children and young people born after the introduction of the hepatitis vaccine within six weeks of birth: 2002 in Benin and 2005 in Senegal.

The types of vaccine used in each population were also not explored, in order to compare the relative efficacy of each type of vaccine.

5. Conclusion

Our study involved cohorts of predominantly female populations recruited in Cotonou and Dakar. The rate of protection against hepatitis B virus, although low in both populations, was higher in Dakar than in Cotonou. This rate is improving in our populations. Awareness of the disease remains low. Vaccination policies and the dissemination of knowledge about the viral hepatitis B epidemic need to be strengthened.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

- [1] Fattovich, G., Bortolotti, F. and Donato, F. (2008) Natural History of Chronic Hepatitis B: Special Emphasis on Disease Progression and Prognostic Factors. *Journal of Hepatology*, **48**, 335-352. <https://doi.org/10.1016/j.jhep.2007.11.011>
- [2] WHO (2020) Hepatitis B Key Facts. <https://www.who.int/news-room/fact-sheets/detail/hepatitis-b.2022>
- [3] World Health Organisation (WHO) (2020) Principaux repères sur l'hépatite B. <https://www.who.int/fr/news-room/fact-sheets/detail/hepatitis-b>
- [4] Programme National de Lutte contre les Hépatites (PNLH) Sénégal (2018) LE PLAN STRATÉGIQUE NATIONAL. <http://hepatites.sn/index.php/combattre-les-hepatites/la-reponse-du-senegal>
- [5] OMS. Le Bénin en bonne voie pour l'élimination des hépatites virales d'ici l'horizon 2030. <https://www.afro.who.int/fr/countries/benin/news/le-benin-en-bonne-voie-pour-lel-elimination-des-hepatites-virales-dici-lhorizon-2030>
- [6] Laouina, A., *et al.* (2016) Évaluation analytique de la méthode chimioluminescence (CMIA-Architect i 2000 SR) pour la qualification biologique des dons de sang en virologie. *Transfusion Clinique et Biologique*, **23**, 310. <https://doi.org/10.1016/j.tracli.2016.08.119>
- [7] Ahmed Ben, A., *et al.* (2004) La demande de soins dans les centres de médecine générale du Sahel tunisien. *Cahiers d'études et de recherches francophones/Santé*, **14**, 223-229.
- [8] Soliman, G., *et al.* (2019) Mass Screening for Hepatitis B and C in Southern Upper Egypt. *BMC Public Health*, **19**, Article Number: 1326. <https://doi.org/10.1186/s12889-019-7640-1>
- [9] WHO (2017) Vaccins anti-hépatite B. <https://iris.who.int/bitstream/handle/10665/255873/WER9227-369-392.pdf;jsessionid=34FD6859D2CEDE36AA0D6BEAEA833A5F?sequence=1>
- [10] Lawson-Ananissoh, L.M., *et al.* (2015) [Knowledge of Hepatitis B Virus among Outpatients Consulting in the Hepatogastroenterology Department at the Lome Campus University Hospital. *Médecine et Santé Tropicales*, **25**, 319-322.

- <https://doi.org/10.1684/mst.2015.0489>
- [11] Okonkwo, U., *et al.* (2018) Public Awareness of Hepatitis B Virus Infection in Cross River State, Nigeria: A Population-Based Survey. *West African Journal of Medicine*, **35**, 79-84.
- [12] Denis, F., *et al.* (2019) [Evaluation of the Immunization Status against Hepatitis B and of the Prevalence of Ag HBs among Beninese Soldiers Participating in Military Missions in Ivory Coast]. *The Pan African Medical Journal*, **32**, 19. <https://doi.org/10.11604/pamj.2019.32.19.16840>
- [13] Maguette Sylla Niang, K.S.F., Babacar Mbengue, Moustapha Mbow, Ndiokhor Nd. Diouf, Ousseynou Boye, Gora Lo, Papa Madieye Guèye, Rena Derwiche, Amina Sow Sall, Tandakha Ndiaye Dieye, Alioune Dieye (2017) Immunological Status to Hepatitis B Virus of Pregnant Women in Dakar, Senegal. *Open Journal of Immunology*, **7**.
- [14] Lo, G., *et al.* (2019) Hepatitis B Virus (HBV) Infection amongst Children in Senegal: Current Prevalence and Seroprotection Level. *The Pan African Medical Journal*, **32**, 140. <https://doi.org/10.11604/pamj.2019.32.140.14485>
- [15] Kodjoh, N. (2015) [Fighting Viral Hepatitis B and C in Africa. Focus on Benin]. *Médecine et Santé Tropicales*, **25**, 141-144. <https://doi.org/10.1684/mst.2015.0439>