



# Seroprevalence of Hepatitis B Virus Infection (HBsAg) in Rural Blood Donors, Moba, Tanganyika Province, Democratic Republic of Congo (2014 to 2016)

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## Abstract

Blood transfusion is a salutary practice in modern medicine, but it carries a high risk of transmission of transfusion transmitted infections (TTIs), especially in developing countries. The objective of this study is to determine the seroprevalence of viral hepatitis B among blood donors. This is a retrospective and descriptive cross-sectional study of the period from 2014 to 2016 at the Katele Health Reference Center (Moba, DR. Congo). We investigated HBsAg in blood donors by using Determine<sup>®</sup> HBSAg. 1145 blood donors with an age mean of  $30.6 \pm 6.9$  years and predominantly male (62.5%) were retained. The seroprevalence of hepatitis B infection from 2014 to 2016 was 3.9% (0.3% for 2014, 9.4% for 2015 and 0.7% for 2016). The family blood donors (83.2%) were the only carriers of HBsAg and were significantly associated with hepatitis B infection ( $p < 0.05$ ). Age [OR = 0.70 CI 95% 0.34 - 1.44;  $p$  0.338] and sex [OR = 0.72 95% CI 0.27 - 1.97;  $p$  0.529] were not associated with the occurrence of hepatitis B in blood donors. The risk of transmission of HBV during transfusion remains high. We recommend the strengthening of transfusion safety measures, the abandonment of family donors for regular voluntary donors, the improvement of screening and diagnostic tests, the involvement of the national blood transfusion program in epidemiological surveillance and the mobilization of the population in favor of the fight against hepatitis B.

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## Subject Areas

Hematology, Public Health

## Keywords

Blood Donors, DRC, HBsAg, Hepatitis, Transfusion Safety

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## 1. Introduction

Blood transfusion is a salutary practice of modern medicine for the management of severe anemias that urgently need blood [1]. Blood transfusion has been used for more than eighty years and presents risks of transmission of infectious agents transmitted by transfusion [1]-[8]. These agents include the human immunodeficiency virus (HIV), hepatitis B virus (HBV), hepatitis C virus (HCV), hepatitis E virus, *Treponema pallidum*, *Trypanosoma cruzi* and *brucei* (*T. brucei brucei* and *T. brucei gambiense*), Cytomegalovirus, Human T lymphocyte virus, etc. [4] [5] [6] [8] [9] [10] [11]. In this study, research is being conducted on the hepatitis B virus (hepatotropic Hepadnaviridae family) in blood donors.

Viral hepatitis B (HVB) is an inflammation of the liver with impairment of hepatocytes that degenerate following infection with hepatitis B virus (HBV) [2] [12]. It is a serious public health problem [2] [12] [13] [14] and patients are at risk of liver cirrhosis and hepatocarcinoma [2] [7] [10] [12] [13] which can lead to hepatic encephalopathy, due to insufficiency Hepatocellular, and death [2] [9] [10] [13]. Around 240 million people are infected with chronic viral hepatitis and more than six hundred and eighty-six thousand individuals die each year as a result of HVB and its consequences [2]. East Asia and sub-Saharan Africa regions are the most affected [2] [15]. HBV is endemic with a different prevalence in different regions of the world [13]. The seroprevalence of markers of hepatitis B virus infection is an indicator that has been variously appreciated among blood donors worldwide: 20% in Tanzania [16]; in Nigeria 14.0% in 2000-2013 [17] and 11.1% in Kano [18]; 10.01% in Equatorial Guinea [10]; 10.0% in Cameroon [19]; 4.7% in Ethiopia [20]; 2.8% in Rwanda [21]; 1.2% in Nepal [22]; 1.1% in India [1] and 0.6% in Namibia [23], etc.

In the Democratic Republic of Congo, the seroprevalence of hepatitis B virus infection among blood donors is 1.6% to 9.2%: Kinshasa 9.2% [24], Lubumbashi 8.0% [25] and 6.8% [6], Kisangani 6.0% [26], Bukavu 3.7% [27] and 1.6% in rural areas of Kamina [28]. In Moba, there are no studies evaluating the seroprevalence of hepatitis B (carriage of HBS antigen) among blood donors. The objective of this study is to determine the seroprevalence of hepatitis B (HBsAg) among blood donors at the Katele Health Center in Moba. It will allow future researchers (local and non-local) and the national blood transfusion program (NBTP) to have a reference in the epidemiological follow-up of HBV infections.

## 2. Materials and Methods

### 2.1. Site, Type and Study Periods

This is a cross-sectional retrospective and descriptive study carried out at the Katele Reference Center in Moba from 2014 to 2016, *i.e.* three years. The Katele Reference Health Center (RHC) is located in Moba-Port, in the health area of Moba-Port (population 2017: 25,266 inhabitants), mainly on the shores of Tanganyika Lake, in the health zone of Moba, Province of Tanganyika in the DRC. It covers an estimated population of more than 60 thousand inhabitants, mainly from health areas along Tanganyika Lake (Regezza, Mulunguzi, Kansenge, Liombe, etc.) over a distance of more than 70 km. The main diseases encountered are, in order of importance: malaria, acute respiratory infections, simple diarrhea, dysentery (especially in artisanal mineral extraction areas), measles, cholera (seasonal pattern) and sexually transmitted infections (STI).

The territory of Moba has an area of 24,500 km<sup>2</sup> and 609,406 inhabitants, *i.e.* a density of 24.9 inhabitants/km<sup>2</sup> [29]. The ethnic populations are predominantly Tabwa (over 80%), Bemba and Luba. There are also other tribes from the DRC and neighboring countries (Tanzania and Zambia). The main activities include agriculture and fisheries. This fishery on Tanganyika Lake attracts people from several towns in the DRC and neighboring countries: Kalemie, Mbuji-Mayi, Kananga, Uvira, Bukavu, Lubumbashi, Kolwezi, Pweto and Kirando (Tanzania). This fishery concerns the species specific to Tanganyika Lake, namely *Stolothrissa tanganyikae* (locally called “DAGA”) and *Luciolates* (locally called “MUKEBUKA”).

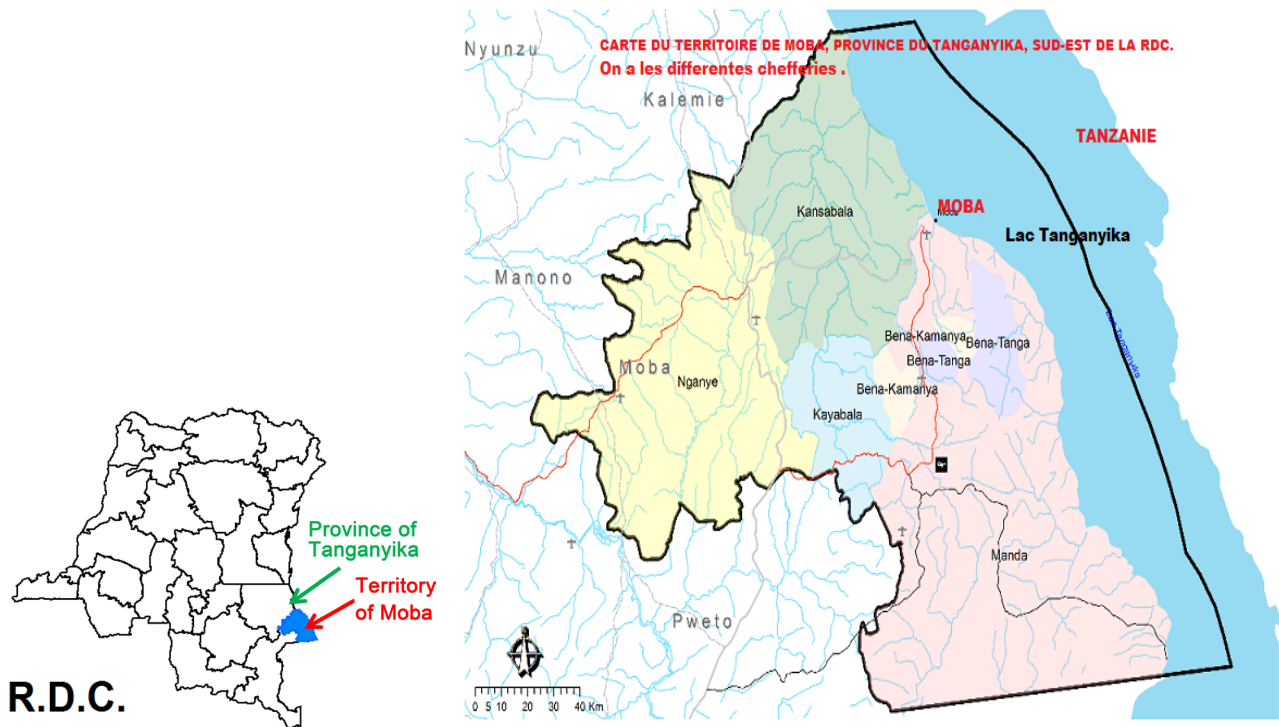
### 2.2. Population and Parameters Studied

Our study population consisted of all blood donors of the period concerned (n = 1145). The age, sex, type of blood donation (family, irregular volunteer circumstantial or paid) and the result of the serological investigation of the HBS Antigen were the desired parameters.

### 2.3. Method of Determination of Viral Hepatitis B

The Australian antigen or HBsAg corresponds to the surface antigen of HBV, discovered accidentally by Blumberg in 1964 [13]. It is currently the most widely used serological marker for the diagnosis of acute and chronic HBV infections (presence of HBS Antigen in serum indicates active, acute or chronic viral hepatitis B) and for the screening of blood donors and organs. The title of circulating HBsAg would reflect the amount of DNA-ccc (covalently closed circular deoxyribonucleic acid) present in the liver [13]. HBsAg is the serological marker appearing 1 to 3 months after the contamination and 2 to 4 weeks before alanine aminotransferases (ALAT) [13] [22].

In the context of this study series, the Rapid Diagnostic Test Determine<sup>®</sup> HBsAg (Abbot, Tokyo, Japan) was used to research for HBsAg according to WHO guidelines [2] [30]. Determine<sup>®</sup> HBsAg is an immunochromatographic test for the qualitative detection of hepatitis B surface antigen (HBsAg). The blood sample is deposited on the sample deposition area and migrates to the deposition area of



**Figure 1.** Geolocation and boundaries of the Moba territory. Moba is one of the five territories of the Tanganyika Province (Moba, Nyunzu, Kongolo, Manono and Kabalo). He is located in the South-East of Democratic Republic of Congo. Source: Map from [www.caid.cd](http://www.caid.cd).

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the conjugate; it is reconstituted and mixed with the selenium-antibody colloid conjugate **Figure 1**. This mixture continues to migrate on the solid phase to the immobilized antibodies at the patient window on the test. If HBsAg is present in the sample, it binds to the antibody of the selenium antibody-colloid conjugate and the patient window antibody by forming a red line. If, on the other hand, HBsAg is absent, the selenium antibody-colloid conjugate passes through the patient window without forming a red line. A procedure control bar is included in this assay system to ensure the validity of the assay [13] [29] [31]. The laboratory at the Katele HRC uses a single HVB (Determine<sup>®</sup> HBsAg) screening test without confirmation by more sensitive and specific tests such as Enzyme-Linked Immunosorbent Assay (ELISA).

#### 2.4. Data Processing and Analysis

The data collected on a pre-established form were encoded on the Excel table (Microsoft, USA, 2010) before being exported for processing on the Epi Info 7.1 software (CDC, USA, 2012). The results were presented in the form of tables and figures showing observed numbers, frequencies, proportions and parameters of central tendency and dispersion (mean, standard deviation, median as necessary). The association between the variables studied was assessed using the unadjusted Pearson chi-square test. The allowed alpha error was 5% and any value of  $p \leq 0.05$  was considered significant.

### 3. Results

The results of this study concern the 1145 blood donors registered during the study period at the Katele Health Reference Center (Moba, DRC) laboratory. Of the 1145 blood donors examined, 926 (80.9%) were between 18 and 38 years of age. The mean age was  $30.6 \pm 6.9$  years with extremes ranging from 18 to 49 years (**Table 1**). Men were the most represented among blood donors followed by women with respectively 715 (62.5%) and 430 (37.5%). In terms of blood donor categories (types), family donors were the most commonly encountered: 953 (83.2%), followed by paid donors ( $n = 112$  or 9.8%) and irregular volunteers ( $n = 80$  or 7.0%). The proportion of voluntary blood donations was lowest and only concerns circumstantial irregular voluntary donors (**Table 1**).

The study of the prevalence of viral hepatitis B (HBsAg) in our study series revealed a seroprevalence of 3.9% ( $n = 45$ ) over three years (**Figure 2**). The year 2015 was the most contributory with 9.4% ( $n = 42$  out of 448 registered blood donors). The years 2014 and 2016 had very low seroprevalence with respectively 0.3% ( $n = 1$  of 393 donors) and 0.7% ( $n = 2$  of 304 blood donors). Throughout the study period, cases of HBV infection were only registered in the family donor category, *i.e.* 100%. HBsAg seropositivity cases (HBsAg carriers) had an average age of  $29.6 \pm 7.2$  years and extremes of 19 - 42 years. The seronegatives had an average age of  $31.1 \pm 6.8$  years and the extremes of 18 to 49 years (**Table 1**).

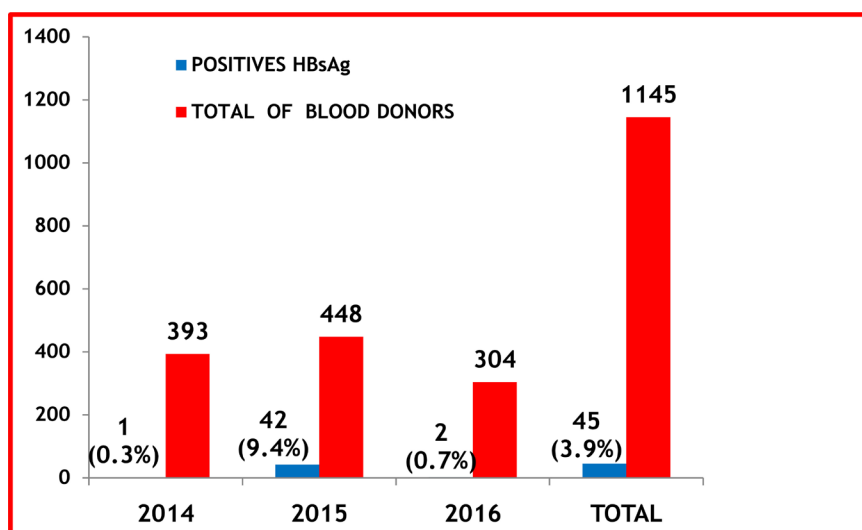
Among the carriers of HBsAg, the female ( $n = 19$  or 4.4%) and the 18 to 38 years age group were most significantly affected ( $p > 0.05$ ).

Family donors were significantly associated with hepatitis B infection ( $p < 0.05$ ). In our study series, age [OR 0.70 CI 95% 0.34 - 1.44] and sex [OR 0.72 CI 95% 0.27 - 1.97] were not associated with the occurrence of hepatitis B in blood donors ( $p > 0.05$ ) (**Table 2**).

**Table 1.** Characteristics of blood donors and seroprevalence of HBV.

Characteristics	Total n (%)	Seropositive n (%)	Seronegative n (%)
Age (years)			
18 - 38	926 (80.9)	39 (3.4)	887 (77.5)
39 - 58	219 (19.1)	6 (0.5)	203 (18.6)
Means	30.6	29.6	31.1
Sd	6.9	7.2	6.8
Mediane	30.5	29	30
Extremes	18 - 49	19 - 42	18 - 49
Sex			
Male	715 (62.5)	26 (2.3)	689 (60.2)
Female	430 (37.5)	19 (1.7)	411 (35.9)
Types of donors			
FD	953 (83.2)	45 (3.9)	908 (79.3)
VD (=CIVD)	80 (7.0)	0	80 (7.0)
PD	112 (9.8)	0	112 (9.8)

Sd: Standard Deviation; FD: Family Donors; PD: Paid Donors; VD: Volunteer Donors (=Circumstantial irregular volunteer donors); CIVD: Circumstantial Irregular Volunteer Donors.



**Figure 2.** Evolution of the prevalence of HBV infection from 2014 to 2016.

**Table 2.** Characteristic analysis and seroprevalence of HBV (HBsAg) in blood donors.

Variables	Total n (%)	HBV (AgHBS) to the blood donors			p-value
		Seropositive n (%)	Seronegative n (%)	OR (CI 95%)	
Age (years)					
18 - 38	926 (100)	39 (4.2)	887 (95.8)	0.7 [0.34 - 1.44]	0.338
39 - 58	219 (100)	6 (2.7)	213 (97.3)		
Sex					
Male	715 (100)	26 (3.6)	689 (96.4)	0.72 [0.27 - 1.97]	0.341
Female	430 (100)	19 (4.4)	411 (95.6)		
Types of donors					
FD	953 (100)	45 (4.7)	908 (95.3)		
VD (=CIVD)	80 (100)	-	80 (100)		0.032
PD	112 (100)	-	112 (100)		

OR: Odd Ratio; IC Confidence Interval; Sd: Standard Deviation; FD: Family Donors; PD: Paid Donors; VD: Volunteer Donors (=Circumstantial Irregular Volunteer Donors); CIVD: Circumstantial Irregular Volunteer Donors.

## 4. Discussion

### 4.1. Characteristics of Blood Donors

During the period of our study on the prevalence of HBV infection in blood donors, blood donors were found to be young adults aged 18 to 49 years and an average of  $30.6 \pm 6.9$  years. This age corresponds or is close to what some authors [2] [20] [24] [25] [32] [33] [34] [35] [36] had encountered in different countries. At this age of intense physical activity, individuals consider themselves strong and able to give blood as old men, already weakened by the weight of the age. Male individuals were the most registered among blood donors (62.5%). This

observation has been preferentially evoked in several studies [1] [5] [14] [20] [22] [24] [25] [28] [37] [38]. For Batina *et al.* [26], blood donors were mainly women. The arguments favoring male predominance take into account the cultural habits and certain physiological predispositions of women to be excluded from the category of blood donors. Indeed, on the one hand to give blood, the donor must feel stronger and fit for that responsibility. In Africa, it is the man who is considered to be the strongest and responsible for any situation that threatens the family: therefore it is better able to give blood than the woman. On the other hand, pregnancy and breastfeeding are contraindications to blood donation, which would reduce women's chances of donating blood. Some authors also mention menstrual blood loss in women [5] [6]. Blood donors were predominantly family donors (83.2%) [5] [25]. This study demonstrates the persistence of paid and family donors in our environment. These categories of donors have been associated for years and in several regions of the world with the risk of transmission of infectious agents during blood transfusions [12] [15] [24] [25] [26] [28] [32] [33] [38] [39] [40] [41] [42]. We believe that the trust of family members in family blood donation and the poor public mobilization policy in favor of regular blood donations are the basis of this situation [8]. Health authorities and partners involved in blood safety and management should be involved to resolve this situation as in some parts of the world and the Democratic Republic of Congo [27] [28] [33] [43]. Our safety of the blood recipient must not remain only in the embryonic stage dating back to 2001 (rehabilitation and inauguration of the National Center for Blood Transfusion) [44]. Several awareness campaigns and funding of national blood transfusion program (NBTP) activities for voluntary and regular blood donations are therefore necessary.

#### 4.2. The Seroprevalence of HBV Infection in Blood Donors

In Africa, the prevalence of HBV infection is 0.2% to 20% among blood donors [5] [10]. The cumulative seroprevalence of HBV infection (HBsAg) during the three years of study was 3.9%. It is close to the results encountered by Kabinda [33] 4.2% and Namululi [27] 3.7% in the DRC, and Rakotoniaina [14] in Madagascar with 3.21%.

This prevalence of HBV among blood donors is lower than in some studies: 20% in Tanzania [16], 14.96% in Burkina Faso [45], 14.0% in Nigeria [17], 4.7% in Ethiopia [20] and 8.8 in Uganda [21]. However, it is higher than India's 1.1% [1], in Nepal 1.2% [22], in Canada 0.007% to 0.06% [46], in Iran 0.15% [47], China 1.085% [48], Italy 0.007 [49] and Rwanda 2.8% [21].

In the Democratic Republic of Congo, this seroprevalence is lower than Kaki-singi [25] 8.01% and Michel [6] 6.8% in Lubumbashi, Batina in Kisangani 6.0% [7], Mbendi in Kinshasa 9.2% [24]; greater than 1.6% obtained by Kabamba Nzaji [28] in rural Kamina; and close to 3.7% of Namululi [27] and Kabinda [33] 4.2% of volunteer donors in Bukavu. Infectious risk in blood recipients appears to be certain in all provinces of the DRC and viral hepatitis B continues to be the reason for exclusion from donating blood. This risk is more acute in developing



countries than in developed countries [47]. The difference between countries and certain environments relative to others is related to socioeconomic status, the level of education of the population, the organization of the health system, the vaccination status of the population and the quality of selection Of candidates for blood donation [47] [50].

The time-series trend in the evolution of the seroprevalence of HBV infection in our study does not make it possible to give a serious and accurate judgment on the evolution of HBV infection in our environment. The peak was observed in 2015 with 9.4% while 0.3% in 2014 and 0.7% in 2106. It would be necessary to wait for the evolution of this seroprevalence in the following years to make the judgment. However, we believe that on the whole, this fluctuating behavior would be influenced by family donors known as donors at risk of transfusion-transmitted infection (TTIs) [32] [40] [41] [51].

In our study, the observed seroprevalence of HBV infection in females (4.4%) was higher than that of males (3.6%). The difference observed is not statistically significant ( $p > 0.05$ ). This was mentioned by Michel [6] in the DRC. Similarly, the prevalence of HBSAg in blood donors aged 18 to 38 (4.2%) and 39 to 58 years (2.7%) was not statistically significant ( $p > 0.05$ ). Unlike Xie [10], for who age was associated with the high seroprevalence of HBV infection in Equatorial Guinea. On the other hand, the blood donation category is associated with HBV infection.

Family donors were the only ones affected by HBV in our series and the difference observed was significant ( $p < 0.05$ ) [6]. It is here that the need for NBTP to organize public awareness campaigns in favor of regular voluntary blood donations in order to achieve the WHO 2020 targets: 100% voluntary donations [52].

For this study, age and sex are not statistically associated with the occurrence of Hepatitis B infection. Michel [6] and Batina [7] fund the significative association between age and the AgHBSAg in Lubumbashi and Kisangani (DRC). The rural environment has been identified as associated with the risk of transmission of hepatitis B by Kabinda [33], which requires the strengthening of epidemiological surveillance measures for blood donors.

### **4.3. From the Method of Screening and Diagnosis of HBV Infection: Qualitative Assay of HBsAg**

Apart from the risk of contamination of the blood recipient during the transfusion discussed in this work, it is useful to highlight the limits of our strategy of securing blood donations. Indeed, in addition to the fact that the NBTP recommends the use of a single rapid diagnostic test (RDT) to assess the risk of HBV infection, Determine HBsAg tests are recognized to be less sensitive in some studies [53] [54] [55]. The ELISA chain can be considered as an alternative for confirmation of the results [25] [26] [34]. In Madagascar, in the CHUA-JRA Ampeloha Immunology laboratory, the results obtained during the evaluation of the rapid tests for the detection of HBsAg showed a sensitivity of 96.1%, a specificity



of 93.2%, 93.6% PPV (positive predictive value), 95.8% NPV (negative predictive value) for the Determine HBsAg<sup>®</sup> Kit, whereas optimal diagnostic criteria require sensitivity and specificity greater than 98% [55].

A study conducted in India [56] between 2004 and 2005 revealed that 1027 cases of hepatitis B surface negative antigen were positive in 18% antibody to HBV antibodies and 21% with DNA-HVB. In our study series, no serological results were confirmed, which may limit the scope of our work.

Determine HBsAg<sup>®</sup> is a qualitative, in vitro, visual-quality immunoassay for the detection of hepatitis B surface antigen (HBsAg) in human serum, plasma or whole blood [13] [57] [58]. No test can absolutely guarantee that a sample does not contain low concentrations of HBsAg, such as those presented at a very early stage of infection. Therefore, a negative result does not exclude the possibility of exposure to HBsAg or infection with HBsAg [31] [34] [57] [58].

In order to increase our ability to diagnose and distinguish acute HBV infection from chronic infection in endemic Congolese we recommend: (1) That the NBTP uses two RDTs to diagnose HBsAg (instead of a single test currently performed): we are a low-income country and the cost of HBsAg research by RDTs would be affordable for all Structures; (2) Long-term use of ELISA [23] [34]. In the absence of ELISA, detection of HBsAg may be associated with the patient's symptoms and other hepatitis B viral serum markers such as anti-HBc, anti-HBc and viral DNA (There are low cost automata) depending on whether or not they have been vaccinated [24] [57] [59]. Deoxyribonucleic acid (DNA) from HBV can be detected two to three weeks prior to the detection of HBsAg [60]. For Blanco *et al.* [61] in Argentina, the nucleic acid test contributes greatly in reducing the potential infectious risks of transmission of hepatitis B virus.

Anti-HBc is the first antibody to appear in serum following exposure to HBV, less than one month after the onset of HBsAg. It is not recommended for the detection of hepatitis B but is a useful marker in the context of acute hepatitis B diagnosis in endemic areas [24] [59]. In Malaysia, one thousand HBsAg negative subjects were found to be 87.3% positive for Anti-HBc by ELISA [43]. However in our context, no paraclinic examination is very necessary than the better selection of blood donor candidates (pre-test counseling).

Securing blood donations is a responsibility of the government [50] and its partners who should make better supplies of good quality blood to the Congolese population. This includes regular voluntary donations, good quality pre-test counseling, more effective screening and diagnostic tests, training of blood transfusion personnel and the availability of sufficient blood banks and epidemiological follow-up Seroprevalence in both urban and rural areas.

## 5. Conclusion

The results of our study demonstrate the need for epidemiological monitoring of blood donations and the strengthening of transfusion safety measures. The overall seroprevalence of HBsAg of 3.9% over three years was influenced by the highest peak observed in 2015 (9.4%), while prevalences of 0.3% and 0.7% were observed

in 2014 and 2016. The instability of the seroprevalence of hepatitis B virus infection among blood donors and family donors is insufficient to ensure healthy blood donation among recipients in our environment. Apart from the negative impact of family blood donations, we should highlight the limitations of TDR (Determine<sup>®</sup> HBsAg) used in our context with respect to HBV sensitivity and specificity. The NBTP should incorporate more efficient tests into its strategy and, if necessary, a second diagnostic confirmation test. Safekeeping of the recipient should not remain in the embryonic stage dating back to 2001 (rehabilitation and inauguration of the National Blood Transfusion Center). The new strategies should be adapted according to field research data.

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## Conflict of Interest

The authors do not declare any conflicts of interest in connection with this study.

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