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# **Infectious Diseases and Epidemiology**

RESEARCH ARTICLE

# Prevalence of Haemoparasites among Blood Donors Attending the Regional Hospital Center of Franceville (Southern Gabon)

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#### **Abstract**

**Background:** Blood donation is a lifesaving intervention and a precious gift for patients in urgent need. However, the blood should be carefully checked for transfusion-transmitted infections such as HIV, hepatitis, syphilis, malaria and other endemic haemoparasites. In Gabon, like in many other African countries, blood is not routinely screened for malaria as recommended by the World health organization.

**Objective:** The main objective of this study was to screen the blood of prospective donors attending Amissa Bongo Regional Hospital Center of Franceville (Southern Gabon) for blood-borne parasites.

**Methods:** Blood donor candidates were subjected to a structured questionnaire and haemoparasites detected using microscopic examinations: direct examination and cytoconcentration technique for filarial worms, thick blood smears and direct examination for *Babasia spp* and *Trypanosoma spp*, SD Bioline Malaria AG P.F/PAN Test and thick/thin blood smears for *Plasmodium falciparum*.

**Results:** The majority of blood donor candidates were males, family donors, work in the informal sector, and under 39 years old. The results also revealed that the prevalences of malaria parasites (*Plasmodium falciparum*) and filarial worms (*Loa loa* and *Mansonella perstans*) were respectively 5.59% (51/447) and 2.68% (12/447). In contrast, none of the

prospective blood donors tested positive for Babesiosis or African Trypanosomiaisis. Family blood donors were more affected with malaria compare voluntary donors (OR = 1.45; 95% CI = 0.42 - 5.00). The age group 18-28 was statistically most affected by the filarial infection than the other groups (OR = 8.83; 95% CI = 1.09 - 71.35; P-value = 0.0411). The results also showed that more than one third of prospective donors suffered from anemia. However, there was no significant association with either malaria or microfilaria (P-value > 0.05).

**Conclusion:** The current hospital- based study detected *Plasmodium falciparum* and filarial worms among prospective blood donors. This finding that raises concerns should draw the attention of local health authorities and lead them to routinely screen blood for malaria parasites as recommended by the world health organization.

#### **Keywords**

Haemoparasites, Anemia, Blood donors

#### Introduction

Blood donation is the most precious gift that one can give to a person in urgent need. It is estimated that 112.5 million people donated whole blood Worldwide, and 5.6 million in Africa [1]. However, the safety of blood must be checked for transfusion transmissible infections (TTI)



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namely human immunodeficiency virus (HIV), hepatitis Band C, syphilis, and blood parasites such as *Plasmodium* spp, [2,3]. Another transfusion transmissible parasitic infection that The WHO recommend to be screened for in Latin America is the American Trypanosomiaisis, also known as Chagas disease (CD). The burden of the disease has been substantially reduced due to control initiatives and currently, about 6 to 7 million people suffer from Chagas diseases and 70 million are at risk of infection worldwide [4,5]. The disease is caused by *Trypanosoma* cruzi and primarily transmitted by infected Triatomine bugs. However, few cases of transfusion-transmitted CD have been reported in the literature [6,7]. Human African Trypanosomiaisis (HAT) or sleeping sickness, a different form of trypanosomiasis, is transmitted by the bites of infected tsetse flies infected with Trypanosoma brucei gambiense and T.b. rhodesiense. The former one accounts for 95-97 % of all reported HAT cases, and 5-3% for the latter one; together they put at risk the lives of 54 million people living in the endemic regions [8,9]. The number of HAT cases have significantly decreased in the past decades, from more than 25,000 in 1998 to 663 in 2020 due to control initiatives [10]. HAT is endemic to Gabon and prevalent in seven (7) out the nine (9) provinces of the country [11]. A total of 116 cases have been reported between 2011 and 2020 [10].

Babesiosis is an emerging zoonosis that infects about 10 million people around the World [12]. The disease, caused by *Babesia spp* and transmitted by Ixodesticks, can lead to death in immune-compromised and elderly patients [13]. More than 165 confirmed cases of transfusion-transmitted babesiosis have been reported worldwide in the literature [12,14]. Human babesiosis has been rarely reported in Africa. Only few cases have been described in South Africa, Mozambique, Egypt, and Equatorial Guinea, one the neighboring of Gabon [15]. More recently, a study conducted in Gabon has reported the detection of *Babesia spp* among livestock imported from Cameroon, a neighboring country [16].

Another transfusion transmissible infection is filariasis. The infection is transmitted by arthropod vectors and caused by various microfilaria responsible for causing lymphatic, subcutaneous, and body cavity filariasis [17-19]. Loiasis and Mansonellosis are two filarial infections caused by *Loa loa* and *Mansonella perstans*, respectively. Both infections are transmitted by arthropods, *Chrysops* for the former and *Culicoides* for the latter, and are endemic in 10-33 sub-Saharan Africa countries including Gabon [17,19]. Cases of transfusion-transmitted filariasis are rare and were reported in Chad, India, and the United States with no serious adverse events [20].

According to the World Health Organization (WHO) report published in 2017, the total deferral rates reported by 128 countries in the world, including 30 African countries, varied from less than 1% to over 37%.

A previous global database on blood estimated that 13 million blood donor candidates were deferred due to various health conditions including anemia, medical conditions or the risk of transfusion transmissible infections [21]. Transfusion transmitted malaria is one the most prevalent Transfusion transmissible infection and the first one to have been reported [22], followed by viral hepatitis and HIV [23,24]. More than 3,000 cases of TTM, mostly from non-endemic countries, were reported worldwide [25]. Despite the WHO recommendations, the blood is still not routinely screened for malaria in many African countries including Gabon [1,26], a situation that puts at risk naive blood receivers. Indeed, transfusion-transmissible malaria can cause serious health problems especially in pediatric patients and even lead to death if the diagnosis is delayed [27,28]. Moreover, the latest WHO report based on 85 endemic countries revealed that malaria cases and deaths have increased from 227 million and 558,000 respectively, in 2019, to 241 million and 627,000 in 2020 [29]. The observed increases were mainly caused by the disruption of health services during the Covid 19 pandemic. Therefore, it is critical that the blood be screened for malaria and other endemic haemoparasites to prevent any complication. The main objective of this study was to assess the prevalence of blood borne parasitosis endemic to Gabon among prospective blood donors attending the Amissa Bongo Regional Hospital Center of Franceville, Southern Gabon.

#### **Material & Methods**

### Study design

This cross-sectional study was conducted at the Amissa Bongo Regional Hospital Center of Franceville (Southern Gabon) from September 13<sup>th</sup> to October 30<sup>th</sup> of the year 2021. This hospital center is the largest one of the region.

## Study population

All prospective blood donors attending hospital during the study period were subjected to a structured questionnaire containing socio-demographic information, history of blood donation, and risk factors associated with TTI.

### Malaria diagnosis

The detection of malaria parasites was carried out using three different techniques: SD Bioline Malaria AG P.F/PAN Test (SD Bioline, Gyeonggi-do, Republic of Korea), Thick and thin blood smears. All blood samples were first screened with the malaria rapid diagnostic test, positive samples were then confirmed by the Thick blood smears method [30], and the Plasmodium species was identified using the thin blood smears method.

#### Microfilaria detection

The detection of microfilaria was done using two

different techniques: the examination of wet smear (direct examination) and the cytoconcentration technique.

**Direct examination:** Ten (10) µl of fresh blood was placed on a clean and grease-free glass slide using a calibrated micropipette. The drop of blood was covered with a cover slip. The preparation was read at the 10 x objective after waiting for 1 minute. Microfilariae are often found at the margin of the blood drop.

Cytoconcentration technique: The cytoconcentration technique was done as described by Petithory and collaborators [31]. Five (5) ml of blood collected on EDTA tube were centrifuged for 5 minutes at 2,000 rpm and the supernatant discarded. Next, the pellet was resuspended in 2 ml of 0.9% physiological water and lacquered for 10 minutes. The suspension was centrifuged for 10 minutes at 2,000 rpm and the supernatant discarded. Next, the pellet was resuspended in 1 ml of 2% saponin and centrifuged for another 10 minutes at 2,000 rpm. The supernatant was discarded, the pellet resuspended in 3 ml of 0.9% physiological water, and centrifuged at 2,000 rpm for an additional 10 minutes. Then, a drop of the pellet was placed in the center of a clean and degreased glass slide using a Pasteur pipette. The preparation was covered with a cover slip, and the slide read at the 10 x objective after waiting for 1 minute.

# **Babesiosis and African trypanosomiaisis**

Babesiosis was detected by thick blood smears and African trypanosomiaisis by both thick blood smears and direct examination of fresh blood.

### Data management and statistical analysis

All participants were subjected to a standard blood donor questionnaire containing information about socio-demographic data, blood donor history, and potential risk factors before providing a blood sample. Blood samples were tested for various blood borne parasites and biochemical parameters. The generated data were enter into Excel spread sheets, exported and analyzed using MedCalc® Statistical Software version 20.027 (MedCalc Software Ltd. Ostend. Belgium; https://www.medcalc.org; 2022). The characteristics of the study population were described using descriptive statistics. ODDS ratio and Pearson's Chi square test used to determine the degree of association between the disease and independent variables. A P-value ≤ 0.05 was considered statistically significant.

# **Ethical consideration**

All prospective blood donors who signed an inform consent were included in the study. Participants' identities were kept secret and permission was obtained from the hospital internal review board. The study protocol was conducted in line with ethics rules contained in the Declaration of Helsinki.

### **Results**

# Socio-demographic characteristics and clinical history of prospective blood donors

A total of 447 prospective blood donors (51 female and 396 male) were included in the study (Table 1). The median age of participants was 30.45 ± 7.75 with an age ranging from 18 to 50. The majority (374) of the study participants were family donors and nearly 85% (379/447) had donated blood previously. Among all prospective donors, 161 (36%) declared to sleep under a bed net and 210 (47%) to use insecticide sprays. The questionnaires also revealed that 72 (16%) and 43 (9.6%) participants had a previous episodes of malaria and microfilaria, respectively. The majority of participants work in the informal sector (151) and participants under 39 years were the most prevalent age group (354).

**Table 1:** Socio-demographic characteristics and clinical history of prospective blood donors attending the Amissa Bongo Regional Hospital Center, Southern Gabon.

Variable	Frequency (%)			
Gender				
Female	51 (11.40)			
Male	396 (88.60)			
Age				
18-28	172 (38.50)			
29-38	182 (40.70)			
39-50	93 (20.80)			
Occupation				
Student	115 (25.72)			
Unemployed	76 (17)			
Formal	105 (23.50)			
Informal	151 (33.78)			
Previous malaria infection				
Yes	72 (16.10)			
No	375 (83.90)			
Use of Bed net				
Yes	161 (36)			
No	286 (64)			
Use of Insecticide Spray				
Yes	210 (47)			
No	237 (53)			
Previous microfilaria				
infection				
Yes	43 (9.62)			
No	404 (90.38)			
Type of donors				
Voluntary donor	73 (16.33)			
Family donor	374 (83.67)			
Donated blood previously				
Yes	379 (84.78)			
No	68 (15.22)			

**Table 2:** Association of malaria infection with independent variables of blood donors attending the Amissa Bongo Regional Hospital Center, Southeastern Gabon.

Variable	Frequency (%)	Malaria + (%)	Odd ratio	95% CI	P-value
Gender					
Female	51 (11.40)	1 (1.96)		0.43 - 24.36	0.2563
Male	396 (88.60)	24 (6.06)	3.22	0.40 - 24.00	0.2000
Age					
18-28	172 (38.50)	10 (0.58)			
29-38	182 (40.70)	12 (0.66)	1.14	0.48 - 2.72	0.7616
39-50	93 (20.80)	3 (3.22)	0.54	0.14 - 2.01	0.3587
Occupation					
Student	115 (25.73)	7 (6.08)	1.57	0.39 - 6.29	0.5190
Unemployed	76 (17)	3 (3.95)			
Formal	105 (23.50)	6 (5.71)	1.47	0.36 - 6.09	0.5914
Informal	151 (33.78)	9 (5.96)	1.54	0.40 - 5.87	0.5253
Previous malaria infection					
Yes		4 (5.55)			
No	72 (16.10)	21 (5.6)	0.99	0.33 - 2.97	0.9880
	375 (83.90)				
Use of Bed net	161 (36)	11 (6.83)			
Yes No	286 (64)	14 (4.9)	0.70	0.31 - 1.58	0.3943
	200 (04)	14 (4.9)			
Use of Insecticide Spray					
Yes	210 (47)	40 (0 40)			
No	237 (53)	13 (6.19)	0.00	0.26 4.04	0.0053
Type of donor		12 (5.06)	0.80	0.36 - 1.81	0.6053
Voluntary donor	73 (16 22)	3(4.1)			
•	73 (16.33)	22(5.88)	1.45	0.42 - 5.00	0.54
Family donor	374 (83.67)	, ,			
History of blood					
donation					
Repeat donor	319 (71.36)	18(5.64)			
First time donor	128 (28.64)	7(5.46)	1.03	0.42 - 2.54	0.9423

# Prevalences of blood borne parasites among prospective donors

Donors were screened for blood borne infections endemic to Gabon, namely Malaria, Filariasis, and African trypanosomiaisis. In addition, Babesiosis, an emerging disease, was also investigated due to reported cases imported livestock. None of the participants tested positive to either babesiosis or African trypanosomiaisis. However, the prevalences of malaria and filariasis were respectively 5.59% (51/447) and 2.68% (12/447) with only one female testing positive in each case (Table 2 and Table 3). All 25 malaria positive samples were *Plasmodium falciparum*. Among microfilaria positive samples, seven (7) were *Loaloa* and five (5) *Mansonella perstans* giving prevalences of 1.56 % and 1.12%, respectively (Table 4).

# Association of malaria and independent variables

Table 2 results showed that there were no associations between malaria and the age group, the occupation, previous malaria episode, use of bed net or insecticide, history of blood donation. Male were three (3) times more likely to test positive to malaria than female participants; However, the difference was not statistically significant. Likewise, family donors were more prone to tested positive to malaria (OR = 1.45; 95% CI =0.42 - 5.00) compare to voluntary donors, the statistical difference was not significant (P-value = 0.54).

# Association of microfilaria and independent variables

The results of Table 3 showed that there were no significant associations between microfilaria and the

**Table 3:** Association of microfilaria infection with independent variables of blood donor candidate attending the Amissa Bongo Regional Hospital Center, Southeastern Gabon.

Variable	Frequency (%)	Microfilaria + (%)	Odd ratio	95% CI	P-value
Gender					
Female	51 (11.40)	1 (1.96)			
Male	396 (88.60)	11 (2.77)	1.43	0.18 - 11.30	0.7354
Age					
18-28	172 (38.50)	8 (4.65)	8.83	1.09 - 71.35	0.0411
29-38	182 (40.70)	1 (0.55)			
39-50	93 (20.80)	3 (3.22)	6.03	0.62 - 58.82	0.1219
Occupation					
Student	115 (25.73)	2 (1.74)			
Unemployed	76 (17)	2 (2.63)	1.53	0.21 - 11.07	0.6755
Formal	105 (23.50)	2 (1.90)	1.09	0.15 - 7.93	0.9268
Informal	151 (33.78)	6 (3.97)	2.33	0.46 - 11.80	0.3039
Previous microfilaria infection?					
Yes	43 (9.62)	2 (4.65)	1.92	0.41 - 9.07	0.4093
No	404 (90.38)	10 (2.47)			
Often stay in village?					
Yes	207 (46.31)	7 (3.38)	1.64	0.51 - 5.26	0.4016
No	240 (53.69)	5 (2.08)			
Type of donor					
Voluntary donor	73 (16.33)	2 (2.74)	1.02	0.22 - 4.77	0.9746
Family donor	374 (83.67)	10 (2.67)			
History of blood donation					
Repeat donor	319 (71.36)	6 (1.88)			
First time donor	128 (28.64)	6 (4.68)	1.81	0.56 - 5.81	0.3180

**Table 4:** Prevalence of haemoparasites among prospective blood donors attending the Amissa Bongo Regional Hospital Center, Southern Gabon.

Haemoparasites	Positive (%)
Plasmodium spp.	25 (5.59)
Trypanosoma spp	0 (0)
Filarial worms	12 (2.68)
Loa loa	7 (1.56)
Mansonella perstans	5 (1.12)
Co-infection	1 (0.22)
Babesia spp.	0 (0)
Total	37 (8.27)

gender, the occupation, previous episode of microfilaria, regular trips to villages, donor type, blood donation history. However, the age groups 18-28 and 39-50 were respectively 8.8 and 6 times more likely to test positive to microfilaria. The age group 18-28 was statistically most associated with the filarial infection than the other groups (P = 0.0411).

# Prevalence of anemia among blood donor candidates

The overall prevalence of anemia was 34.45 % (154/447) among prospective blood donors (Table 5). Malaria and microfilaria positive blood candidates were more susceptible to suffer from anemia than negative ones. However, neither the malaria nor the microfilaria status was significantly associated with anemia. Likewise, gender, age, occupation, type of donor, and history of blood donation was not significantly affected the prevalence of anemia.

### **Discussion**

The main objective of the study was to determine the prevalence of blood borne parasites (Plasmodium, Microfilaria, Babesia, and Trypanosoma) among prospective blood donors attending the Amissa Bongo Regional Hospital Center in Franceville (Gabon) between September 13<sup>th</sup> and October 30<sup>th</sup> of the year 2021. The results revealed that 88.59 (396/447) of blood donor candidates were males. This result is similar to the prevalences of 83% (83/100) and 96.10 % (296/308) reported respectively by Adusei, et al. [32] in Ghana, and Olawumi, et al. [33] in Nigeria, and consistent with the worldwide general trend [21]. The data also showed that over 15% (68/447) of the participants were first

**Table 5:** Prevalence of anemia among blood donor candidates attending the Amissa Bongo Regional Hospital Center, Southeastern Gabon

Variable	Frequency (%)	Anemia (%)	Odd ratio	95% CI	P-value
Gender					
Female	51 (11.40)	23(45.09)	1.66	0.92 - 2.99	0.0916
Male	396 (88.60)	131(33.08)			
Age					
18-28	172 (38.50)	58(33.2)			
29-38	182 (40.70)	64(35.16)	1.066	0.68 - 1.65	0.7751
39-50	93 (20.80)	32(34.40)	1.03	0.60 - 1.75	0.9102
Occupation					
Student	115 (25.73)	40(34.78)	1.16	0.66 - 2.04	0.5978
Unemployed	76 (17)	26(34.21)	1.13	0.60 - 2.12	0.6936
Formal	105 (23.50)	33(31.43)			
Informal	151 (33.78)	55(36.42)	1.25	0.74 - 2.12	0.4082
Type of donor					
Voluntary donor	73 (16.33)	24(32.87)	1.08		
Family donor	374 (83.67)	130(34.75)	1.00	0.64 - 1.85	0.7569
<b>Donated Previously</b>					
Repeat donor	319 (71.36)	114(35.74)	1.22	0.79 - 1.89	0.3672
First time donor	128 (28.64)	40(31.25)			
Malaria status					
Positive	25	11 (44)	1.53	0.68 - 3.46	0.3043
Negative	422	143 (33.88)			
Microfilaria status					
Positive	12	6 (50)	1.94	0.62 - 6.11	0.2585
Negative	435	148 (34.02)			

time blood donors. This finding is lower than the 58.4% (180/308) reported [33] in Nigeria. However, family donors made up the majority of blood donors in both studies with respectively 83.67 (374/447) in Gabon and 92.7%) (286/308) in Nigeria. This result is consistent with the 2011 -WHO report that revealed less than 25% of voluntary blood donors in Gabon [21]. The reluctance of participants to donate blood voluntarily could be due to cultural believes, ignorance and the willingness to give blood to relatives in distress. Socio-demographic data also revealed that the majority of blood donors were employed in the public and private sectors (57.28%). This finding is in line with percentages of 46.4 and 52 reported in previous studies in Nigeria and Ethiopia [33,34].

Malaria and microfilariasis prevalences were respectively 5.59% and 2.68 %; only one case of coinfection was recorded (Table 4). In contrast, none of the candidates tested positive to either babesiosis or African trypanosomiaisis. Regarding babesiosis, no case of human infection has yet been reported in the country. This could be explained by the difficulty of microscopically and clinically distinguishing malaria

from babesiosis [35]. Moreover, the recent detection of *Babesia spp* and *Boophilus decoloratus* ticks in livestock imported to Gabon from Cameroon raises concerns for humans [16,36]. The absence of African trypanosomiaisis is consistent with the fact that the Estuaire province not the Haut - Ogooue province is the most active site [11]. These negative results could also be explained by the small size of the study sample.

Plasmodium falciparum was the only plasmodium species detected among donors with an overall prevalence of 5.59%. This is consistent with the fact that *P. falciparum* is the major plasmodium species in Gabon [29] and similar to the 4.1 % (17/416) reported by Alemu and Mama in Ethiopia [34]. Malaria prevalences by gender were 2 % and 6% for females and males, respectively; but, the difference was not statistically significant (Table 2). This result is very similar the study conducted in Nigeria by Uneke [37], and consistent with the fact that male donors made up nearly 90% of all participants in both studies. Family donors were more susceptible to Malaria infection compared to voluntary donors (OR = 1.45; 95% CI = 0.42 - 5.00; P = 0.54); but there no statistical difference between the two groups.

Our finding is in line with a study conducted in India that reported a higher malaria infection rate among family donors [38]. In contrast, in Nigeria, Olawumi and collaborators have reported slightly higher malaria prevalence in voluntary donors (28.6 %) compare to family donors (26.6 %); however, the difference was not statistically significant [33]. This discrepancy could be explained by the difference in the adherence to the preventive measures between the two groups or type of occupation. Voluntary donors tend to adopt safer behavior as they often donate blood; and thus, are aware of risky behaviors unlike family donor who are coerced to give blood.

Microfilaria worms were detected in 2.68% (12/447) of prospective donors; of which 1.56 % (n = 7) were Loa loa and 1.12% (n = 5) Mansonella perstans (Table 2). This result is similar to the rates of (1.04% -1.33 %) and (0.33% -0.58%) reported respectively for Loa loa and M. perstans in two separate studies conducted in Nigeria [39,40]. Among all the variables examined in this study, only the age group 18-28 was significantly associated microfilaria (OR = 8.83; 95% CI = 1.09 - 71.35; P-value < 0.05). This finding is in line with that of Bola et al. who reported the highest microfilaria prevalence in the age group 21-30 [39]. This could be explained by the fact that the majority (5/8) of the responders who tested positive falls within this age range and have reported to frequently stay in the villages. Moreover, according to a large scale study conducted in eleven (11) endemic countries in Africa using the Rapid Assessment Procedure for Loiasis (RAPLOA) revealed a prevalence of eye worm history greater than 40% in all Gabonese villages (65) surveyed across the country [41]. In addition, all neighborhoods in the study area are surrounded by vegetation, and thus constitute potential reservoirs of Chrysops or Culicoides. The presence of microfilaria in donors' blood is of matter of concern as transfusion-transmitted filariasis could cause allergic reactions in blood recipients [20].

Regarding anemia, the prevalence among blood candidates in the current study (34%, 154/447)was higher than those 21% and 13.7% reported in Nigeria [42,43]; but, slightly lower than the rate of 36.5% (141/386) reported in Democratic Republic of Congo [44]. Various diseases and conditions could account for the observed differences including but not limited to malaria, iron deficiency, haemoglobinopathies and hemolytic anemia [45,46]. Females were more affected by anemia compare to male donors, most likely due to menstruations.

#### Conclusion

The current study reports the detection of haemoparasites, namely *Plasmodium falciparum* and microfilaria of *Loa loa* and *Mansonella perstans*, among prospective blood donors attending the Amissa Bongo Regional Hospital Center of Franceville, Southern

Gabon. This is the first study, in Gabon, to ever report malaria cases among blood donor candidates to the best of our knowledge. The prevalence of malaria parasites in blood candidates raises concerns as transfusion-transmitted malaria can lead serious adverse events especially in immunocompromised patients or citizens from non-endemic countries. In addition, the overall of malaria reported here may be underestimated due to submicroscopic parasites. Therefore, blood bags should be screened for malaria, and other endemics haemoparasites prior to transfusion as recommended by the WHO especially for immune-compromised recipients.

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