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Microfilariae Prevalence and its Association with Anemia Among First-time Blood Donors in Lambaréné, Gabon

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Background: Anemia remains a significant public health concern in Gabon, particularly among children, adolescents, and females. Gabon is also home to two major species of filarial worms, Loa and Mansonella spp., which cause microfilaremia. The epidemiological nexus between hemoglobin (Hb) concentrations and microfilaremia in Gabonese first-time blood donors remains unknown.

Aims: This study aimed to understand better the epidemiological relationship between anemia and microfilaremia to improve donor selection and management protocols.

Study Design: This retrospective and analytical study was conducted among first-time blood donors in Lambaréné between March 2018 and October 2019.

Methods: Participants aged 16-65 years old and weighing a minimum of 50 kg were enrolled using standard donor selection criteria. An automatic hematological analyzer was used to quantify Hb concentrations, and microscopy techniques were used to detect the presence of microfilariae.

Results: Microfilariae were found in 4.8% (35/723) of the 723 first-time blood donors from Lambaréné. Anemia was classified as mild in 35.5% (257/723) and moderate in 1% (7/723). No significant associations were found between the distribution of microfilariae and variables such as age, sex, socioprofessional classification, marital status, or residence. Blood group O donors had a higher prevalence of microfilariae (6%) than non-O donors (2.7%). However, the observed difference was not statistically significant (AOR =2.3, p = 0.052). Furthermore, microfilariae were associated with increased moderate anemia (3.7% vs. 29%, AOR =15.6, p = 0.003).

Conclusion: Our findings highlight microfilaremia as a possible etiological cause of anemia among Gabonese blood donors, emphasizing the need for further research and a potential review of donor management strategies.

INTRODUCTION

Anemia, a global public health issue, disproportionately affects people in low-income and developing countries.[1,2] The endemicity of infectious diseases such as malaria in Sub-Saharan Africa has been associated with increased prevalence rates of anemia, particularly among vulnerable groups, such as children, pregnant women, and nonpregnant women.[3] Furthermore, diseases associated with anemia are prominent, such as microfilaremia, characterized by filarial parasites in the bloodstream. These filarial parasites, which are primarily transmitted by mosquitos, cause various diseases, including lymphatic filariasis, loasis, and onchocerciasis. Their pathogenesis involves lymphatic trafficking, resulting in obstructions, tissue damage, and clinical symptoms such as swelling and inflammation.[4] *Loa* and *Mansonella perstans* emerged as the most common filarial species in Gabon. Despite rare outbreaks of onchocerciasis in the country's southern regions, these two species dominate the epidemiological landscape. Different geographical distributions have been observed, with *Loa* preferring forested areas of Gabon and *Mansonella perstans*, which are more prevalent along the coast.[5] Anemia has a significant impact in Gabon, with prevalence rates of 69.5% in children and 56.7% in adolescents reported, with rates reaching 60.6% in women.[6-8] While Libreville has reported 69.4% and 45% anemia prevalence rates, respectively, among female and male first-time blood donors,[9] data for Lambaréné remains limited. Simultaneously, a study found that the prevalence rate of *Loa* was 26.4%, *Mansonella perstans* was 14.6%, and coinfections were 5.3% in the general Gabonese population.[5]

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This study investigates whether there is a relationship between anemia and microfilaremia in first-time blood donors from a microfilaremia-prone region of Gabon (Lambaréné). Thus, we aim to improve the epidemiological understanding of these diseases and provide insights for better donor selection and management.

MATERIALS AND METHODS

Ethical approval

This study was performed in accordance with the International Conference on Harmonization's Guidelines for Good Clinical Practice, the Helsinki Declaration, and all applicable national and international standards. Before being included in this study, each participant or the participant's legal guardian under 18 years old provided written consent.

Blood donors

A retrospective analysis was conducted from March 2018 to October 2019, encompassing male and female donors, with a significant portion being family or replacement donors (FRDs). A predonation questionnaire was completed by prospective donors. Those aged 16–65 years old and weighing at least 50 kg were eligible. Exclusions were made for pregnant women, recent transfusion recipients, people with clinical manifestations of jaundice, viral hepatitis, infections, or those involved in risky behaviors in the 6 months preceding the donation. Sociodemographic data were collected, and venous blood samples were obtained following standard protocols.

Determination of hemoglobin levels and screening for microfilariae

Hemoglobin (Hb) levels were measured using the DH36 Auto Hematology Analyzer (Shenzhen Dymind Biotechnology Co., Ltd., Republic of China). This was facilitated by a 4-mL venous blood sample obtained at the elbow into an EDTA-containing tube. Donors with Hb levels below 120 g/l (women) and 130 g/l (men) were classified as anemic. After leucoconcentration, a microscopic evaluation was performed to screen for diurnal microfilariae (*Loa* and *Mansonella* spp.). Erythrocytes were lysed in a conical tube with 2% saponin for 2 min before centrifugation at 2,000 rpm for 10 min. The supernatant was decanted, and a 20-µl pellet sample for microscopy (objective x10) was prepared. A double-blind confirmation of the presence of microfilariae was performed, with a third-party consultation in the event of conflicting results. Microfilariae were identified morphologically using Giemsa-stained smears.

Statistical analysis

This study investigates the prevalence of microfilariae and anemia among blood donors, considering various demographic factors, including gender, age, occupation, and donor type.

A univariate logistic regression analysis was conducted to assess the impact of these demographic factors on the prevalence of microfilariae and anemia. Subsequently, the multiple regression model included variables that showed statistical significance. The final results of this analysis provide odds ratios and 95% confidence intervals.

For the statistical analysis, R software version 4.2.1 was used. The level of significance was set at $p < 0.05$.

RESULTS

Sociodemographic characteristics of the blood donors

Among the 723 first-time blood donors recruited, 632 (87.4%) were male, primarily between the ages of 20 and 39 years, accounting for 353 (48.8%) and 206 (28.3%), respectively, with those 50 and older accounting for only 18 (2.5%). Most (715, 98.9%) were FRDs (Table 1). When examining socioprofessional backgrounds, 154 (21.3%) were pupils or students, whereas the least represented were 16 health professionals (2.2%) and 20 administrative staff (2.8%). Notably, 432 (59.8%) donors were single, with 487 (67.4%) residing in Lambaréné's second arrondissement. Only five (0.7%) came from other places. Regarding blood type, the O blood group was the most common (464, 64.2%), and a significant 703 (97.2%) were Rhesus positive. In contrast, non-O blood groups and Rhesus-negative donors comprised 259 (35.8%) and 20 (2.8%) donors, respectively (Table 1).

Prevalence of microfilariae and anemia in blood donors

Of the 723 first-time blood donors examined, 4.8% (35/723) tested positive for microfilariae, particularly from the species *Loa* and/or *Mansonella* spp. Regarding anemia, 35.5% (257/723) had mild anemia, whereas 1% (7/723) had moderate anemia (Table 1).

Morphological identification of the microfilariae

Microscopic characteristics used to identify microfilariae were size, presence or absence of sheath, headspace, and tip.

Loa species had a large size, a visible sheath, a short headspace with regular nuclei, and a pigtailed tip with thin nuclei extending to the tip, whereas *Mansonella* spp. had a small size, a headspace with irregular nuclei, and a thick nucleus at the tip.

Prevalence of microfilariae based on the sociodemographic characteristics of blood donors

We assessed the prevalence of microfilariae (*Loa* and *Mansonella* spp.) in first-time blood donors based on the sociodemographic characteristics shown in Table 1. This study found that males had a slightly higher occurrence of microfilariae than females, with percentages of 5.1% and 3.3%, respectively. However, this difference was not statistically significant ($p = 0.466$). When the prevalence of microfilariae was examined across different age groups, it was found that those aged 20-29 years had the lowest occurrence at 4.2%. However, there were no significant differences in the bivariate analysis when this group was compared with the 30-39 age group (Table 1). Technical workers had the highest infection rate of any profession (7.4%), but the differences within professional categories were insignificant. Interestingly, cohabiting donors were less likely to have microfilariae than married donors, with prevalence rates of 4.2% and 9.7%, respectively, although this difference was not statistically significant ($p = 0.193$). The residential area of the donors had no discernible effect on the prevalence of microfilariae (Table 1).

TABLE 1. Univariate Logistic Regression Analysis of the Prevalence of Microfilariae According to the Sociodemographic Characteristics of Blood Donors.

Variable	N	Positive (%)	COR	95% CI	***p* value**
Gender					
Female	91	3 (3.3%)	Referent		
Male	632	32 (5.1%)	1.6	0.55-6.61	0.466
Age groups (yr)					
< 20	41	3 (7.3%)	2	0.41-7.1	0.338
20-29	353	15 (4.2%)	1.1	0.47-2.77	0.834
30-39	206	8 (3.9%)	Referent		
40-49	105	7 (8.7%)	1.8	0.60-5.06	0.284
≥ 50	18	2 (11%)	3.1	0.44-13.7	0.175
Blood donors					
VNRD	8	0 (0%)	NA	NA	0.987
FRD	715	7 (6.7%)	Referent		
Occupation					
Carriers	58	2 (3.4%)	1.2	0.12-27.5	0.858
Technical activities	121	9 (7.4%)	2.8	0.50-52.8	0.335
Mining, forestry, and oil activities	81	4 (4.9%)	1.8	0.26-36.3	0.599
Commercial and hotel activities	38	2 (5.3%)	1.9	0.18-43	0.594
Teachers	17	1 (5.9%)	2.2	0.08-57.7	0.588
Pupils and students	154	5 (3.2%)	1.2	0.18-22.9	0.885
Personal health	16	0 (0%)	NA	NA	0.993
Defense and security forces	36	1 (2.8%)	Referent		
Administrative agent	20	0 (0%)	NA	NA	0.992
Unemployed	143	8 (5.6%)	2.1	0.36-39.1	0.498
Others	39	3 (7.7%)	2.9	0.35-60.5	0.364
Marital status					
Married	31	3 (9.7%)	2.4	0.53-8.33	0.193
Single	432	21 (4.9%)	1.2	0.56-2.53	0.702
Concubinage	260	11 (4.2%)	Referent		
Residence					
Second district of Lambaréné	487	24 (4.9%)	1.5	0.42-9.34	0.602
First district of Lambaréné	115	6 (5.2%)	1.6	0.35-10.9	0.589
Road to Mouila	59	2 (3.4%)	Referent		
Road to Libreville	44	3 (6.8%)	2.1	0.33-16.4	0.432
Road to Ndjole	13	0 (0%)	NA	NA	0.99
Others	5	0 (0%)	NA	NA	0.994
Blood type					
O	464	28 (6%)	2.3	1.05-5.82	0.051
Non-O	259	7 (2.7%)	Referent		
Rhesus					
Positive	703	35 (5%)	Referent		
Negative	20	0 (0%)			0.987
Hemoglobin level					
Normal	459	17 (3.7%)	Referent		
Mild anemia	257	16 (6.2%)	1.7	0.85-3.49	0.127
Moderate anemia	7	2 (29%)	10.4	1.42-52.3	0.007

95% CI, 95% confidence interval; COR, crude odds ratio; VNRD; voluntary non-remunerated donors.

Blood group O donors had a significantly higher prevalence of microfilariae (6%) than non-O donors (2.7%). However, the observed difference was not statistically significant (AOR = 2.3, p = 0.052; Table 2). Furthermore, the presence of microfilaremia was associated with a higher prevalence of moderate anemia (3.7% vs. 29%, AOR = 15.6, p = 0.003; Table 2).

DISCUSSION

Our findings support the previously reported male predominance (87.4%) among first-time blood donors not only in Gabon[10,11] but also in other Sub-Saharan African regions.[12,13] Individuals aged 20-29 and 30-39 years contributed the most to our donor pool, accounting for 77.1% of the total, reflecting the youthful trend of blood donors in Africa.[14] The Georges Rawiri Regional Hospital largely depends on FRDs, mirroring patterns observed in the southeastern Gabonese locale of Koula-Moutou.[15] This finding is consistent with the existing literature, which shows that family donors are the most common, accounting for more than 75% of the donor pool in Sub-Saharan Africa.[16]

Our study found anemia in 36.5% of first-time donors, with mild and moderate anemia accounting for 35.5% and 1%, respectively. According to studies from rural Kenya,[17] socioeconomic and environmental factors may shed light on this increased prevalence. Microfilariae were found in 4.8% of first-time donors, which is lower than the percentages reported for *Loa* and *Mansonella* spp. in the province of Moyen-Ogooué, with Lambaréné as its capital.[5] Moreover, Diéki et al.[18] confirmed these findings in cities throughout the Haut-Ogooué province.

TABLE 2. Multivariate Logistic Regression Analysis of Microfilariae Prevalence According to Sociodemographic Characteristics of Blood Donors.

Variable	AOR	95% CI	*p* value
Gender			
Female	Referent		
Male	1.9	0.63-8.41	0.314
Age groups (yr)			
< 20	2.3	0.47-8.91	0.247
20-29	1.2	0.48-3.03	0.74
30-39	Referent		
40-49	1.9	0.62-5.71	0.247
≥ 50	3.5	0.49-16.2	0.14
Marital status			
Married	2.1	0.44-7.94	0.29
Single	1.1	0.50-2.42	0.863
Concubinage	Referent		
Blood type			
O	2.3	1.05-5.92	0.052
Non-O	Referent		
Hemoglobin level			
Normal	Referent		
Mild anemia	1.9	0.94-4.00	0.07
Moderate anemia	15.6	1.98-91.9	0.003

95% CI, 95% confidence interval; AOR, adjusted odds ratio.

The low presence of microfilariae could be attributed to two key factors: a high male participation rate (about 80%) and the majority (89.4%) coming from Lambaréné. Previous research reveals a higher prevalence of microfilariae in females and a lower prevalence in lake-like environments such as Lambaréné compared with forested regions.[5] The diagnostic methodology, which relied heavily on microscopic examination, could have been less sensitive in detecting filariae species, such as *Loa*, which is known to be amicrofilaremic in many carriers.[18-20]

Most sociodemographic factors have no significant correlation with microfilariae prevalence. In addition, in this study, ABO blood groups were not associated with microfilaremia. This contrasts with findings from Nigeria[21] and the Democratic Republic of Congo.[22]

Microfilariae and moderate anemia were found to have a tangible association. An illustrative case from China depicts a male patient with notable anemia and *Loa* microfilariae,[23] emphasizing the relationship. Microfilariae may cause anemia by competing with host cells for essential nutrients, according to research from Cameroon and Tanzania.[24-26] Concurrently, other probable causes of anemia, such as nutritional deficiencies and infections such as malaria, must be considered.[27-29]

In contrast, a previous study on children and young adults in Mali found that microfilariae reduce the prevalence of anemia.[30]

Study limitations

Our study has limitations in terms of participant screening, cohort design, and diagnostic methods, although providing valuable insights. Our conclusions may have inherent limitations due to including a control group and comprehensive diagnostic assessments. Future research should address these shortcomings to understand the relationship between microfilaremia and anemia better.

Our findings indicate a possible relationship between microfilaremia and anemia among Lambaréné blood donors. However, a more thorough study is needed to validate these findings to improve donor health and optimize blood transfusion safety.

Ethics Committee Approval: This study was performed in accordance with the International Conference on Harmonization's Guidelines for Good Clinical Practice, the Helsinki Declaration, and all applicable national and international standards.

Informed Consent: Before being included in this study, each participant or the participant's legal guardian under 18 years old provided written consent.

Data Sharing Statement: The data that support the findings of this study are available from the corresponding author upon reasonable request.

Authorship Contributions: Concept- C.B., S.P.; Design- C.B., S.P.; Data Collection or Processing- S.P.; Analysis or Interpretation- M.S., S.P.; Literature Search- S.P.; Writing- C.B., S.P.

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