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Abstract: This study aimed to determine the prevalence of coinfection with malaria and intestinal parasites and assess its association with anemia in school-aged children from rural and urban settlements in Gabon. This cross-sectional study involved afebrile school children recruited at schools between May and June 2021. Blood and stool samples were collected from participants who provided informed consent to participate in the study. Hemoglobin concentration (Hb) was measured using a HemoCue photometer (HemoCue 201, HemoCue, Angelholm, Sweden). Giemsa-stained blood films were examined to detect malaria parasites and any filarial infections, while the Merthiolate-iodine concentration (MIc) method was used to identify intestinal parasitic infections (IPIs). A total of four hundred and seventy (470) school-aged children were successfully enrolled in this study. The observed prevalence rates were as follows: malaria infection at 69.6%, IPIs at 19.1%, filaria at 5.1%, *Schistosoma* infection at 15.0%, and anemia at 29.0%. Coinfections of malaria with IPIs, filaria, and *Schistosoma* were present in 12.3%, 4.7%, and 6.6% of the children, respectively. Malaria and filaria infections were associated with residing in Lastourville (LTV) city and were also correlated with age, whereas IPIs were associated with male gender and living in the city of LTV. Anemia was linked to malaria infection and was more prevalent among children living in rural areas. The findings of this study indicate that malaria, IPIs, and *Schistosoma* infections continue to pose a significant public health problem in the study area, even though only malaria infection appeared to be associated with anemia. Nevertheless, these results highlight the need for implementing control measures to reduce the rates of malaria, IPIs, filaria, and *Schistosoma*, particularly in Lastourville.

Keywords: malaria infection; co-infections; anemia, children, lastourville, gabon

1. Introduction

Plasmodium and intestinal parasitic infections are significant public health issues in tropical and subtropical settings, particularly in poor communities with inadequate sanitation and hygiene. Given that the distribution of these parasitic diseases spatially overlaps, co-infections in the same individual are common and can result in severe morbimortality. Indeed, *Plasmodium* infection (or malaria) is a

leading cause of death, especially among children. In 2020, WHO recorded approximately 241 million clinical cases and 627,000 deaths from malaria worldwide. Sub-Saharan Africa, accounts for 95% of malaria cases and 96% of malaria deaths, with children accounting for 80% of all malaria deaths[1]. Whereas Intestinal parasitic infections (IPIs), caused either by soil-transmitted helminth (STH), protozoan, or both, are responsible for 450 to 840 million cases worldwide, and the majority are reported in developing countries[2]. Although malaria affects human life negatively, *Plasmodium falciparum* infections manifest through/exhibit heterogeneous outcomes ranging from asymptomatic infection to severe disease, which may rely on the parasite threshold[3, 4] as well as the nutritional status of the host[5]. The WHO defined asymptomatic malaria as the presence of asexual parasites in the blood without symptoms of illness[6]. For Lindblade et *al.*, asymptomatic malaria is the existence of malarial parasitemia of any density in blood without any symptoms in individuals who have not received recent anti-malarial treatment in each population[7]. Whereas, Bousema et *al*. argue that this definition should include early detection of rising parasitemia or any density of parasitized red blood cell (RBC) that is not enough to trigger a fever response[3]. In Gabon, where malaria transmission is perennial, studies have investigated the distribution of asymptomatic malaria in the country. So far, most of the studies conducted in Gabon were cross-sectional studies[8–12] and involved children[10, 12], or adults[9, 11]. They were mostly carried out in rural settings [11, 12]. Nevertheless, one longitudinal survey was conducted on SAC in rural settlements[13]. Besides studies reporting the distribution of asymptomatic malaria in a given population, some surveys have explored the related immune response[8, 14], the genetic diversity of *Plasmodium* sp. in infected individuals with or without sickle cell disease[12], the human genetic polymorphisms and the prevalence and profile of asymptomatic malaria[10], the profile of 10 cytokines in asymptomatic malaria children living in different settings[15]. But there is little data regarding the association between asymptomatic malaria and IPIs. However, a study that investigated the effect of schistosomiasis and STH on the prevalence and incidence of *Plasmodium falciparum* infection highlighted that STH enhances the risk for *Plasmodium* sp. infection in schistosomiasis-positive children and, when infected, that schistosomiasis enhances susceptibility to developing malaria in young children but not in older children[13]. Despite previous studies, our understanding of the epidemiology of this coinfection in Gabon remains limited. There is particularly a lack of data on the coinfection of *Plasmodium* and IPIs in urban areas like Franceville and rural areas like Lastourville. This study aims to assess the prevalence and determinants of asymptomatic malaria and IPIs in schoolchildren in different settlements. The findings of this study could provide valuable insights to Gabonese health authorities for improving current control strategies.

2. Materials and Methods

2.1. Study Sites and Participants

The study was carried out in two regions of Gabon situated in two provinces of the country: Franceville (province of Haut-Ogooue) (1°37′15″S and 13°34′58″E) and Lastourville (province of Ogooué-Lolo) (0°49′S, 12°42′E). Franceville is the third-largest city in Gabon, whereas Lastourville is a rural agglomeration of several villages. Gabon has an equatorial climate which consists of a short dry season (from 15 June to 15 September) and a long rainy season (from 15 September to 15 June). The study involved primary-aged children of both genders living in both regions whose parents, or legal guardians consented to their participation in the study. Children's involvement was voluntary, and only those who had lived at the study sites for at least three months were included. Before the data was collected, the research team visited the study sites to educate the local authorities and residents on the importance, benefits, and protocols of the research. This study is a cross-sectional study of a cohort of school-aged children between the ages of 3 and 17 enrolled in randomly selected elementary schools at both sites. Only children with signed informed consent from a parent or legal guardian were included. The participants underwent interviews and medical examinations before providing blood, feces, and urine. Blood was collected in ethylenediaminetetraacetic acid (EDTA) tubes, and feces collected in clean, well-labeled stool vials at both study sites. Urine samples were

collected only from participants in Lastourville using plastic screw-cap vials. Blood samples were analysed for malaria and filariasis, feces for the presence of STH eggs, and urine for the microscopic detection of *S. hematobium*.

2.2. Blood Sample Examination

2.2.1. Thick Blood Films

For microscopic detection, Thick Blood Smears (TBS) were prepared and used as described elsewhere[16]. After staining with 20% Giemsa for 20 minutes, the slides were examined under a 100x oil-immersion objective. Parasitemia was calculated for all positive TBS, as the number of parasites per microlitre of blood. If no parasites were found after examining 100 oil immersion fields, the slides were considered negative.

2.2.2. Detection of Microfilaria

The detection of microfilaria was carried out using the Sang–Petithory leucoconcentration technique[17]. All microfilaria found on the slide were identified. Due to its higher sensitivity, this technique is indicated when the parasite density is low. Leucoconcentration was applied to all participants who provided blood.

2.3. Fecal Examination by MI Concentration

Fecal samples were analyzed using the Merthiolate-iodine concentration (MIc) method, described by Sapero and Lawless[18], but slightly modified. Briefly, 20 ml of distilled water was added to the plastic screw cap vials containing around 1 g of each sample and homogenized using an applicator stick. 5 ml of this preparation were collected and transferred into a conic tube, then centrifuged for 4 minutes at 3000xrpm. The supernatant was discarded by rapidly inverting the tube, and 500μl of distilled water was added, then mixed well. A drop was collected and placed on a glass slide and blended with a drop of MI solution. Covered, the entire preparation under the cover slide was examined after one minute for eggs, cysts, and larvae using a 10x and 40x lens of a light microscope. Each slide was examined in duplicate by two experienced technicians. Once only one parasite was found, the sample was considered positive.

2.4. Urine Screening for Schistosome Eggs

Urine samples were reviewed using the centrifugation method previously described by [19]. Haematuria was determined either visually (general haematuria) or microscopically (micro haematuria). As stated above, microscopic detection of *S. hematobium* eggs was applied to urine samples obtained from participants in Lastourville only.

2.5. Measuring the Haemoglobin Concentration

Hemoglobin (Hb) concentration was measured using a HemoCueH photometer (HemoCue 201, HemoCue, Angelholm, Sweden). Anemia was defined as a Hb level of <11 g/dl and further classified according to WHO anemia thresholds : severe anemia: Hb <7 g/dl, moderate anemia: Hb 7–9.9 g/dl, mild anemia: Hb 10–10.9 g/dl[20].

2.6. Definitions and Endpoints

Asymptomatic malaria parasitemia was defined as the presence of *Plasmodium* in the blood by microscopic, with an axillary temperature of <37.5°C and no record of fever in the past 2 weeks.
- Parasitemia was categorized as low (<1000 parasites/μl of blood), moderate (1000-4,999 parasites/μl blood), and high (≥5000 parasites/μl blood)**[21]**.

3. Results

3.1. Study Population

A total of four hundred and seventy School Aged Children (SAC) originating from Franceville (Urban) and Lastourville (rural) were included in the study. The general characteristic of the children are summarized in table 1. Overall, there were 249(or 53%) children from urban and 221(or 47%) from rural area. In total, 247 children were females (52.6%), 222 were males (47.4%), with a sex ratio of 0.9. The mean age of the participants was 10.04 (±3.2), and the mean age was higher in rural area than in urban area (p=0.03). Haematological parameters [hemoglobin, white blood cells, red blood cells (Hb, WBC, RBC)] differed significantly between the rural (Lastourville) and urban areas (Franceville) (p <0001). Platelet counts also differed significantly between the rural, and urban areas (p < 0.001).

Table 1. Characteristic of the Study population, according to the study Area.

	Urban n (%)	Rural n (%)	Total
Gender(N=469)			
Male	122(49.2)	100(45.2)	222(47.3)
Female	126(50.8)	121(54.7)	247(52.7)
Total	248(52.9)	221(47.1)	469
Sex ratio	0.96	0.82	0.9
Age group (N=460)			
3-10	138(55.4)	111(44.6)	249(54.1)
11-17	101(47.9)	110(52.1)	211(45.9)
Total	239(51.9)	221(48.1)	460
Parameters			
Mean temperature± SD (°C)	37.0±0.4	36.7±0.8	36.9±0.7
Mean age ± SD (year)	9.7±3.2	10.3±3.1	10.04±3.19
Haemoglobin (g/dl)	11.8±1.3	11.2±1.3	11.66±1.4
WBC (×103/µl)[a]	7.3±2.1	9.3±9.5	8.08 ±5.92
RBC (×106/µl)[b]	4.42±0.5	2.49±4.9	4.01±2.48
Platelet (×103/µl)	294.2±107.1	1983.7±1154.7	838.95±1029.79

3.2. Pattern of Infection Diversity and Prevalence

Three hundred and twenty-seven (327) children were positive for malaria parasites, resulting in a prevalence of 69.6% (95% CI: 65.4-73.7) (table 2). The overall prevalence was higher in rural areas (208/221 [94.1%]) than in urban areas (119/249 [47.8%]). The prevalence of Plasmodium infection was, respectively 71.2% in males and 68.0% in females, irrespective of the location. Malarial parasitemia was higher in children aged between 11 to 17 (162/211 [76.7 %]) compared to children below eleven (163/250 [65.2 %]). The prevalence of low, moderate, and high parasitemia in the study population was 92.7% (303/327), 4.3% (14/327), and 3% (10/327), respectively. The overall prevalence of parasitemia was higher in rural areas (208/324 [64.2 %]) than in urban areas (115/324 [35.5 %]), irrespective of the parasitemia level. The parasite density ranged between 3 and 41450 (mean ± SD = 1021.803 ± 3537.998).

Among the four hundred and seventy participants, only 402 children provided stool samples. 77 children were positive for intestinal parasites, resulting in a prevalence of 19.2% (95% CI: 15.3-23.0). Infection with IPIs was more common in urban areas than in rural areas (24.7% vs 13.3 %). The prevalence of infection was higher in males (47/192 [24.5%]) than in females (30/209 [14.4 %]). Intestinal parasite infections were higher in children aged between 11 and 17 (20.3%) compared to children below eleven (18.3%). *Ascaris lumbricoides* infection was the most prevalent IPI (33 [42.8%]), followed by *Entamoeba coli* (32 [41.5%]), hookworm infections (13 [16.8%]), and whipworm (*Trichuris trachiura*) (8[10.3%]). Infection with *Endolimax nana*, *Taenia saginata*, *Giardia* sp, and *Enteromonas hominis* were the least common IPI identified (at 3.8%, 2.5%, 2.5%, and 1.2%, respectively).

Overall, 24 children had positive TBS for microfilaria infection (5.1%), with 3.4% for *Mansonella perstans* and 1.7% for *Loa loa*. Filarial infection was more common in rural area than in urban area

(10.1% vs 0.8%). The prevalence of filarial infection was, respectively, 5.7% in females and 4.5% in males, irrespective of the location. Regarding age, the prevalence was, respectively, 2.4% for children aged 3 to 10, and 8.6% for those above 10. Concomitant infections with *Loal loa* and *M. perstans* in the study population were not detected.

As stated before, only children from Lastourville were screened for urinary schistosomiasis. Out of the 212 respondents, 15.1% (N=32) of the children had a positive diagnosis for schistosomiasis based on the egg observation by microscopy. Overall, the prevalence was, respectively, 12.8% in females and 17.8% in males. Among the 32 schistosomiasis-positive children, 31 (96.8%) were from the same the place called "Gare de Setrag".

Table 2. Prevalence of Malaria, IPI, Filaria and *Schistosoma* by location, age groups and gender.

		Malaria	**IPI**	**Filaria**	***Schistosoma***
Location					
	Urban n(%)	119(47.8)	51(24.7)	2(0.8)	NA
	Rural n(%)	208(94.1)	26(13.3)	22(10.1)	32(14.5)
Age groups					
	3-10	163(65.5)	38(18.3)	6(2.4)	11(12.2)
	11-17	162(76.8)	38(20.3)	18(8.6)	21(19.4)
Gender					
	Male	158(71.2)	47(24.5)	10(4.5)	17(17.8)
	Female	168(68.0)	30(14.4)	14(5.7)	15(12.8)
Total	**n (%)**	**327(69.6)**	**77(19.2)**	**24(5.1)**	**32(15.1)**

3.3. Plasmodium sp., IPIs, Microfilaria, S. hematobium, and Coinfection Pattern

Globally, the prevalence of co-infections with asymptomatic malaria and IPIs was 12.3%; whereas malaria parasites and microfilaria co-infection sood at 3.6%; and malaria parasites+*Schistosoma hematobium* was 5.3%.

Among the 57 children co-infected with asymptomatic malaria and IPIs, the most frequent combination was *Plasmodium* sp. and *A. lumbricoides* (50.9% or 29/57), followed by *Plasmodium* sp. and *Entamoeba coli* (38.6%, or 22/57), *Plasmodium* sp. and *Ancylostoma* sp. (15.8% or 9/57). A less common association was observed between *Plasmodium* sp. and *Trichuris trichiura* (12.3% or 7/57), while co-infections with *Endolimax nana* were rare, appearing in only 3.5% of cases. Additionally, *Giardia*sp and *Taenia saginata* were each detected alongside *Plasmodium* sp. in a single fecal sample. We noted cases of double parasitism involving *Plasmodium* sp. such as *Plasmodium* sp. plus *A. lumbricoides* (24 cases), *Plasmodium* sp. plus *Entamoeba coli* (15 cases), and less frequently, *Plasmodium* sp. plus *Ancylostoma*sp (5 cases), *Plasmodium* sp. plus *Trichuris trichiura* (2 cases), and *Plasmodium* sp. plus *Endolimax nana* (1 case). A few instances of triple parasitism also emerged: one child presented with a combination of *Plasmodium* sp., *Ancylosyoma*sp, and *Giardia*sp, while another harbored *Plasmodium* sp., *Entamoeaba coli*, *Ancylostoma*sp, and *Taenia saginata*, all in stool samples collected from urban area.

Notably, all children infected with microfilaria parasites were also coinfected with malaria parasites with no cases of combined microfilaria infection, i.e., they either harbored *Plasmodium* sp. plus *Loa loa* or P*lasmodium* sp. plus *Mansonella perstans*.

3.4. Risk Factors Associated with Malaria, IPI, Filaria

Univariate analysis revealed that the risk of malaria infection was significantly associated with age (OR =1.124, 95% CI: 1.054-1.200, p< 0.001). Furthermore, the risk of malaria infection was markedly higher in children dwelling in rural area (OR= 17.627, 95% CI: 9.882-33.996). Interestingly, gaining one kilogram in weight was linked with an increased risk of malaria infection, although this association was marginally significant (p=0.037). The multivariate logistic regression model showed that both age and place of residence (City) were associated with increased risk of malaria infection (table 3).

Table 3. Risk factors associated with malaria in logistic models (simple and multiple).

	Univariate				Multivariate			
		95%CI				95%CI		
	OR			p-value	OR			p-value
		Lower	Upper			Lower	Upper	
Gender (male)	1.161	0.783	1.726	0.459	1.386	0.858	2.254	0.185
Age	1.124	1.054	1.200	**<0.001**	1.037	0.909	1.179	0.587
Weight	1.020	1.002	1.039	**0.037**	1.023	0.987	1.064	0.222
Temperature	1.009	0.991	NA	0.654	1.565	1.081	2.259	**0.015**
City(LTV)	17.627	9.882	33.996	**<0.001**	23.649	12.256	49.767	**<0.001**

A logistic regression model (detailed in table 4) demonstrated that the factors associated with intestinal parasitic infections in the univariate model were similarly significant in the multivariate model. Male gender and residing in the city of LTV emerge as key factors linked to a heightened risk of malaria infection.

For filarial infection, a logistic regression model with Gender, Age, weight, temperature and City as independent variables, indicated that Age and residing in LTV (p<0.001) are significant risk factors for filarial infection. The odds of carrying filarial infection are presented in table 5.

Table 4. Risk factors associated with IPI in logistic models (simple and multiple).

	Univariate				Multivariate			
		95%CI				95%CI		
	OR			p-value	OR			p-value
		Lower	Upper			Lower	Upper	
Gender (male)	1.934	1.170	3.239	**0.011**	1.732	1.028	2.950	**0.040**
Age	1.057	0.976	1.145	0.175	1.183	1.024	1.371	**0.023**
Weight	1.006	0.983	1.028	0.626	0.967	0.926	1.009	0.127
Temperature	0.988		1.010	0.782	0.845	0.588	0.989	0.379
City(LTV)	0.468	0.275	0.780	**0.004**	0.377	0.206	0.672	**0.001**

Table 5. Risk factors associated with filaria in logistic models (simple and multiple).

	Univariate				Multivariate			
		95%CI				95%CI		
	OR			p-value	OR			p-value
		Lower	Upper			Lower	Upper	
Gender (male)	0.789	0.334	1.802	0.577	0.829	0.327	2.043	0.685
Age	1.328	1.149	1.559	**<0.001**	1.384	1.063	1.838	**0.020**
Weight	1.040	1.007	1.073	**0.014**	0.983	0.915	1.050	0.615
Temperature	1.137	1.044	2.292	0.699	1.826	1.038	3.869	0.098
City(LTV)	13.806	4.003	86.835	**<0.001**	14.016	3.786	91.433	**0.001**

3.5. Prevalence and Risk Factor for Anemia

In this study, the overall prevalence of anemia was found to be 29.0% (110/379, 95%CI: 24.43-33.57), with no difference between sexes (p=0.59). The prevalence rates of mild and moderate anemia were 62.7 % (69), and 36.4 % (40), respectively. One chlid had severe anemia (0.9%). The anemia rates were not influenced by the age category (p=0.0246). Moreover, the prevalence of anemia was higher in rural area (41.2%) compared to urban area (22.5%). Among children diagnosed with asymptomatic malaria, a noteworthy 93 (38.1%) were anemic. A significant association was established between anemia and malaria infection (see Table 6).

Table 6. Correlations between different infections and anemia**.**

	IPI	Filaria	Malaria	Anemia status
IPI	1			
Filaria	-0.009	1		
Malaria	0.040	0.110	1	
Anemia status	-0.015	0.030	0.270	1

3.6. Different Parasites and Polyparasitism

Polyparasitism i.e, infection with more than one parasite, was observed in this study. Infection with multiple parasites (blood, stool and/or urine parasites) was prevalent in 24.4% (99/405) of study population. There were twelve different parasites, among which seven helminths (*Ascarislumbricoides*, *Ancylostoma*sp, *Trichuristrichiura*, *Loaloa*, *Mansonellaperstans*, *Taeniasaginata* and *Schistosomahematobium*), and 5 protozoans (*Plasmodium* sp, *Entamoebacoli*, *Endolimaxnana*, *Enteromonas*sp, and *Giardia*sp). The mean number of parasite species per participant was 1.67 (± 1.02). It was higher in respondents living in rural area (2.06±0.78) than in urban area (1.32±1.08) (p=1.0810-15). The mean was, respectively, 1.6(±1.02) and 1.71(±1.02) in females and males. There were more children infected with more than one parasite in the rural area (63.6%) than in the urban area (36.4%) (X^2=94.5, p<2.210-16). Polyparasitism was more common in children aged between 11 to 17 than in those below 11 (58.6% vs 41.4%). Gender had no influence (58.6% vs41.4%, p=0.199). Double parasitism was more prevalent (75/99).

4. Discussion

The present study investigated the co-occurrence of asymptomatic malaria parasites and co-infections in schoolchildren from two different settlements in Gabon. While asymptomatic malaria parasites have been screened in some studies, there is limited data on their co-occurrence with other infections in Gabonese schoolchildren.

In this cross-sectional study, various associations between asymptomatic malaria and other infection were revealed. among the asymptomatic malaria co-infected, 12.3% had IPIs, 4.7% had filarial infections, and 6.7% had *Schistosoma* infection. Previous studies from Gabon have reported the coinfection of malaria with IPIs 7%[22], filaria 0.2%[22], and *Schistosoma* 9%[13] at the infection rates slightly different to those found in the present study. These differences might be due to the target population and the sample size. In most of the previous studies, the association between malaria and other infections was mainly isolated in febrile individuals when compared to afebrile ones with, in some studies, the cohort consisted of 428 febrile vs 88 afebrile[23]; 410 febrile vs 60 afebrile[24]; 793 febrile vs 100 afebrile[25]. Whereas in our study, a cohort of around 470 afebrile children only was screened. Nevertheless, the hypothesis that these discrepancies are confounded by socioeconomic, genetic, and nutritional factors should not be rolled out.

Overall, a high prevalence of asymptomatic malaria in children, was observed at a rate of 69.7%, which is higher than the 57.08% and 42.92% recorded in febrile and afebrile children from Lastourville[26], and the 52% recorded in adults in Lambarene regions[9]. Our findings are also higher than that of the study reported in other African countries such as CAR, 35.2%[27], and Cote d'Ivoire, 50.3%[28]. The difference might be due to limited access to diagnostics, treatment, and prevention during the global covid 2019, or the seasonal variation and the geographical difference of the study populations[29, 30]. This means that these trends of high asymptomatic malaria rates may impair the efforts of local authorities to eliminate malaria. Because these patients may act as reservoirs for the malaria parasite and be involved in autochthonous transmission cycles[31, 32]. Rural areas recorded a higher malaria prevalence and parasitemia than urban areas, probably because of limited access to control strategies[22], environmental factors, and the high entomological inoculation rate[33]. This study confirms the heterogeneity of malaria burden and transmission intensity in Gabon[31]. Older children (11-17 years of age) were more likely than younger children counterparts to be infected with malarial parasites, which is consistent with previous reports[31]. According to

[34], this pattern might be explained by the relationship between age and insecticide-treated mosquito net use. In their study and many others in Africa, children in the younger age group were significantly more likely to sleep under insecticide-treated mosquito nets, which have proven to be highly protective against malaria[34]. However, other studies have reported a decreased prevalence of asymptomatic malaria when the age of the participant increase. This situation was related to the development of protective immunity after cumulative exposure to the parasite and the acquisition of knowledge on malaria prevention and control strategies[30].

In the present study, the prevalence of IPIs was 19.1%, which is lower compared to the 61.1% reported in different settlements of Gabon by[22] , 49.0% in Lambaréné by[35]. Compared to similar studies done in other countries, the prevalence of IPIs was also found to be lower than reported in communities around Buea in Cameroon (47.2%)[36], Nigeria (24%)[37], Angola (44.2%)[38], and Mozambique (31.6%)[39]. Lower prevalence rates have been reported in the hospital in Cameroon (11.9%) [40], Ghana (15%)[41], and Ethiopia (15.5%)[42]. Intestinal parasitic infections were more prevalent in male children than female children, regardless of the location and the parasite species, consistent with findings of other studies[41]. These discrepancies could be attributed to the difference in behavior, males often play outside in more contact with soil than females, who are more involved in the household chores. Among the isolated intestinal parasites, *Ascaris lumbricoides* was the predominant parasite causing infection in children, consistent with what was reported in other areas of Gabon by[22] and elsewhere[37, 38].

The prevalence rates of microfilaria infection, by *L. loa* and *M. perstans*, were almost the same with that reported by[22], but low contrary to other data of previous studies of other areas in Gabon, in which at leats 15% of the screened individuals had one or both of the worms[43, 44]. The microfilariasis rates was more prevalent in rural area than in urban, in line with other studies conducted in other areas of Gabon[22, 45].

Anemia is a significant public health problem for school children in malaria-endemic areas, affecting physical growth, cognition, and academic performance. Although its causes are diverse in tropical areas – ranging from helminths and hemoglobinopathies to malnutrition – the implementation of malaria control measures such as long-lasting insecticide-treated nets (LLINs), artemisinin-based combination therapy (ACTs), indoor residual spraying and mass administration of antihelmintic drugs (MDAs) are public health strategies for the control of anemia in schoolchildren. A high prevalence of anemia (29%) was observed among schoolchildren, around 85% of anemic individual had asymptomatic carriage of *Plasmodium* infection. This prevalence of anemia is in consonance with observations in Cameroon (30.8%[46]), in Ethiopia (41.3%[30]), in Nigeria (34.4% [47]), but lower when compared with the >73.5% observed in febrile Gabonese children[48]. Anemia was significantly more common in children under five years old (p = 0.000), with no notable difference based on gender. Young children are particularly vulnerable to anemia, exacerbated by infections from bacteria, malaria, and intestinal parasites. Our findings showed that there was a significant association between anemia and malaria. This is consistent with previous studies which shown malaria, besides IPI infections to have profound effects on anemia in schoolchildren[49–52].

The findings from the study have implications for managing malaria-related morbidities in the two involved settlements, and possibly in other regions with similar conditions. However, there are limitations: parasitological microscopy cannot detect asymptomatic infections at the submicroscopic level, and relying on single slides for soil-transmitted helminths (STHs) may have biased results due to variability in STH egg excretion, especially in children with low intensity. Moreover, the study design does not support causality assessments between asymptomatic malaria and other causes of anemia.

5. Conclusions

This study highlights the high prevalence of asymptomatic malaria and co-infections with intestinal parasites, filaria, and *Schistosoma* among schoolchildren in two distinct settlements in Gabon. The findings emphasize the silent yet significant burden of these infections, which can contribute to ongoing transmission in endemic regions. Rural areas exhibited higher prevalence rates

compared to urban ones, likely due to disparities in access to control measures and environmental factors. The high rate of asymptomatic carriers, particularly among older children, underscores the need for targeted interventions, as these individuals may serve as reservoirs for further transmission. The co-occurrence of intestinal parasitic infections and microfilariae, as well as the association of malaria with anemia, points to a complex interplay of factors affecting child health in these regions. Anemia, a critical public health issue, was found to be significantly associated with asymptomatic malaria, further stressing the need for integrated strategies to manage both malaria and parasitic infections in school-age populations. The findings of this study underscore the importance of continued surveillance, improved diagnostic techniques, and strengthened prevention efforts to reduce the silent burden of malaria and co-infections in vulnerable populations.

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