Ntoum: A major focus of malaria in Gabon's Estuary province

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Abstract

Introduction: Malaria is a major public health problem in Gabon, and yet in some regions of the country, such as the administrative department of Komo-Mondah, data on malaria are scarce or lacking. The aim of this study was to describe the epidemiological and clinical characteristics of malaria patients in the commune of Ntoum (Komo-Mondah).

Methods: Socio-demographic and clinical characteristics were collected from 248 patients attending the clinical laboratory of Ntoum departmental hospital using a pre-established data collection form. After blood sampling in an EDTA tube, a haemogram and two blood smears (thin and thick) were performed on each blood sample by a laboratory technician.

Results: The prevalence of malaria was 78% (194/248), with *Plasmodium falciparum* identified as the main etiological agent. Those most at risk of malaria were children aged 0-5 years (43%, 83/194) followed by adults older thant 25 years (21%, 41/194), and female (62%, 120/194). Parasitemia ranged from 10 to 210,000 parasites/µL blood, and 73% (140/194) of parasite infections were associated with anemia. Most cases of anemia (83%, 118/140) were mild, 78% (110/140) microcytic anemia of which99 cases were hypochromic anemia.

Conclusion: This study has provided crucial information for understanding the occurrence of malaria and the infectious complications resulting from malaria infection in the Ntoum region. The data also reveal the absolute necessity of implementing targeted and sustainable health education programs for vulnerable groups.

Keywords: Malaria, Plasmodium falciparum, Ntoum, Gabon

Introduction

Malaria is a parasitic disease caused by the intra-erythrocytic multiplication of a haematozoan of the genus Plasmodium, transmitted to man by the bite of a haematophagous female mosquito of the genus Anopheles, which belongs to the Culicidae family, of the order Diptera [1]. This curable disease is particularly rife in intertropical regions, which are ideal for the survival and development of the vector agent [1]. Africa is most affected the region of the world, with 93.6% of cases and 95.4% of deaths in 2022 [2]. The fight against this disease in recent years through the distribution of insecticide-treated mosquito nets and the use of artemisinin-based combination therapy (ACT) has reduced the number of deaths in sub-Saharan Africa by 60% [3].

In Gabon, a territory located in Central Africa, malaria is the leading cause of consultation and hospitalization (24%), with a case-fatality rate of 11 per 100,000 inhabitants [4]. Recent studies on the prevalence of malaria have been carried out in several localities in this country [5, 6, 7]. For several decades, the administrative department of Libreville-Owendo has been the focus of much attention. Numerous epidemiological studies in child health [8, 9], maternal-fetal health [10, 11], and the prevalence of genotypes associated with resistance to certain antimalarial drugs [12, 13] have been carried out there, leaving the other administrative departments of the Estuary province, including Komo-Mondah, unprepared. To the best of our knowledge, no study has been undertaken in Komo-Mondah, and the present observational study aims to describe for the commune of Ntoum: the burden and epidemiological characteristics of malaria, prophylaxis practices applied by the population, and drug prescriptions given to patients suffering from malaria.

Materials and Methods

Study setting and location

This study took place at the departmental hospital in the commune of Ntoum, the only hospital in the region. Ntoum is the capital of the Komo-Mondah department in the Estuary province. Geolocated at 0°23'29" north and 9°45'32" east (Figure 1), the town is 38 kilometers (km) from Libreville, the country's political capital. Situated at the crossroads of access to Owendo, Libreville, Kango and Cocobeach, and close to towns such as Donguila, Bikele and Akok, the town of Ntoum, benefiting from the development of National Road 1, has since experienced record demographic (population estimated at 90,096 in 2013) and urban (48.9 inhabitants/km²) growth [5].

This Gabonese region offers a suitable living environment thanks to the availability of basic public services. The locality enjoys a transitional equatorial climate with a bimodal, austral-type regime. Over the year, the average temperature is 26.3°C and rainfall is abundant, around 3,000 mm/year. Food plantations (bananas, manioc, yams, etc.) intersected by patches of secondary forest occupy a large part of the region. Despite high rainfall, the rivers that flow into the bay are small, discharging volumes of water that correspond to the small surface area of the watersheds. Despite the proximity of the political capital, human encroachment on the Mondah mangroves remains very limited. A few fishermen have set up camps and use wood as fuel for smoking fish [14]. This proximity to the wild flora exposes populations to frequent contact with wild fauna and the pathogens they carry, which are liable to transmit disease.

Type and period of study

This is a descriptive observational study conducted over a 3 month period, from August 18 to October 18, 2023, corresponding to the major dry season.

Study population, inclusion criteria

Patients of both sexes and of all ages were enrolled in the consultation room. Inclusion in the study was based on the following criteria: fever (body temperature > 37.0°C), biological work-up including a positive thick blood drop result for a plasmodial species and a blood count.

Sample size

The cohort size was exhaustive, determined according to Daniel Schwartz's formula ($n=z^2pq/i^2$) where: "n" represents the minimum number of patients to be enrolled; "z" the critical value following the reduced centered normal distribution worth 1.96 for a risk of error α (α=5%); "p", the national prevalence of malaria (16% in the Gabonese republic) [4]; "q" worth 1-p= 0.84; "i", the degree of precision (i=0.05). In order to have a direct impact on the accuracy of the statistics produced, the minimum size of the cohort was determined using Daniel Schwartz's formula, which was 207 patients. However, in order to guarantee greater precision and excellent statistical power, 41 additional patients or 20% of the cohort were added, bringing the final number of patients to 248.

Collection of data and biological material

A nurse interviewed patients who met the inclusion criteria and consented to participate in the study. For adult patients, the nurse carefully filled in the pre-established data collection form, specifying socio- demographic and symptomatological characteristics. For minors, oral consent from the legal guardian was required prior to inclusion in the study. A laboratory technician then performed a venipuncture on each patient to collect venous blood in an EDTA tube (VWR International, France).

Laboratory analysis

The ABX micros 60 (Horiba ABX SAS, France) was used to obtain a haemogram from 10 µL of EDTA blood. The parameters measured were white blood cell (WBC), red blood cell (RBC) and hemoglobin (Hb) counts; mean corpuscular volume (MCV), mean corpuscular hemoglobin content (MCHC) and platelet count (PLT). Anemia has been defined according to the World Health Organization (WHO) classification as Hemoglobin (Hb) concentration below 11 g/dL and categorized as mild ($8 \leq Hb < 11$ g/dL), moderate ($5 \geq Hb < 8$ g/dL), and severe ($Hb < 5$ g/dL) [15, 16]. Microcytosis, defined as an abnormality in erythrocyte size, has been determined according to patient sex and age [17].

Blood parasitology analyses for each sample consisted of a thick smear (Lambaréné method) [18], stained with Giemsa diluted to 20%, to observe malaria parasites and their evolutionary stages, and to determine their parasite density (parasitaemia/µL blood). If the slide was positive, a thin smear, also stained with 20% Giemsa, was taken to identify the infecting plasmodial species. Two Biotechnicians read slides with an immersion objective using a light microscope (Leica DM500). A thick smear was considered negative if no parasites were observed after reading 100 microscopic fields. Conversely, a sample was considered positive if at least one parasite was observed on the 100 microscopic fields scanned. Parasite densities were classified as low (less than 2500 parasites/µL), medium (2500 to 9999 parasites/µL), high (10000 to 19999 parasites/µL), very high (20000 to 100000 parasites/µL), and hyper-high (more than 100000 parasites/µL).

Ethical considerations

The General Management of Ntoum Hospital Departmental approved the study.

Results

Socio-demographic characteristics of the study population. During 3 months, 248 consenting patients were enrolled in the study. They ranged in age from 0 to 78 years, with an

average age of 17.83 years. Over half the patients enrolled (61%, 155/248) were female, and five (5)of them were pregnant (Table 1). Male patients represented 39% (97/248) of the total population. The most representative age range was 0-5 years (39% - 98/248), followed by patients older than 25 (25%, 64/248). The remaining age groups, 16-24 and 6-15, represented 19% (47/248) and 17% (43/248) of enrolled patients, respectively. More than half the patients (68%, 172/248) had a secondary school education. The proportion of pupils/students was 32% (80/248). A large number of patients (82/248, 33%) were unemployed, while 15% (36/248) were self- employed (shopkeeper, driver, etc.) and 14% (35/248) were civil servants (teachers, gendarmes, health personnel, etc.).

Characteristics of the malaria population

Of the 248 patients enrolled, 78% (194/248) were thick drop positive (Table 1), and the parasite identified was the Plasmodium falciparum species. The majority of malaria patients were women, 62% (120/194) of whom three were pregnant. 38% (74/194) of patients were men. The age groups most at risk of malaria were 0-5 years (43%, 83/194) and ≥ 25 years (21%, 41/194). The majority of patients (93%, 181/194) lived in Ntoum. The remaining 4% (7/194) and 3% (6/194) came from the neighboring towns of Kango (54 km away) and Libreville (40 km away), respectively.

Blood counts

Blood counts showed that 27% (53/194) of malaria attacks were non-anemic, while 73% (141/194) of parasitic infections were associated with anemia. Among patients with anemia, 17% (23/141) had moderate anemia, compared with 83% (118/141) with mild anemia (Figure 2, Table 2).

Types of anemia

Table 2 shows the changes in blood tissue composition noted in patients during P. falciparum malaria attacks. Of the 141 cases of anemia diagnosed, 78% (110) were
microcytic, 21% (30) normocytic and 1% (1) macrocytic. In the group of patients diagnosed with microcytic anemia, 90% (99/110) of hypochromia cases were observed in just over half the patients aged 0- 5 years (52%, 57/110). In contrast, similar proportions of normocytic, hypochromic or normochromic anemias were diagnosed in patients of different age groups. These erythrocyte changes may or may not be associated with variations in thrombocyte levels. In patients with mild anemia, 26 cases of thrombocytopenia were detected, compared with 30 cases of thrombocytosis. In patients with moderate anemia, 7 cases of thrombocytopenia were found, compared with 4 cases of thrombocytosis, bringing the total number of patients with thrombocytopenia to 33, and those with thrombocytosis to 34 in cases of malaria infection with anemia. For malarial infections without anemia, the total number of subjects with thrombocytopenia was 14, and 15 for those with thrombocytosis. Hypochromic normocytosis was also recorded in 37 patients.

Type of malaria

Physician consultations and laboratory test results revealed 1% (2/194) plausible cases of severe malaria in a 1-year-old child (convulsive, cough, asthenia) and a 28-year- old young adult (loss of consciousness, dyspnea, asthenia/fever) (Figure 2). Cases of uncomplicated malaria (cough, dizziness/headache, asthenia/fever) were observed in 49% (95/194) of the remaining patients, they were 38, 29, 17, and 11 cases aged 0-5 years, ≥25 years, 6-15 years, and 16-24 years respectively. Cases of moderate malaria characterized by vomiting/diarrhea associated or not with headache/vertigo and/or asthenia/fever were observed in 17% (33/194) of patients including 23, 7, 2, and 1 patents aged respectively 0-5 years, 6-15 years, ≥25 years, and 16-24 years.

Parasitaemia

Observed parasitaemia ranged from 10 to 210000 parasites/µL (p/µL) of blood, with a mean of 4752 p/µL of blood. The majority of patients (75%, 145/194) had low parasite densities, while the remainder (17% (34/194), 5% (9/194), 2% (4/194) and 1% (2/194) had medium, high, very high and hyperhigh parasite densities, respectively. The median parasitaemia by age group revealed that patients aged 0-5 years (83 cases) had a parasitaemia of 8,151 (13-210000) p/µL, much higher than those determined in the other age groups: 1. 044 (13-10000) p/µL in the 33 patients aged 16-24 years, 2,098 (13- 16000) p/µL in the 41 adults (≥25 years), and 2,648 (10-30000) p/µL in the 37 patients aged 6-15 years. The different parasite densities observed (low, medium, high, very high and hyper-high) induced the occurrence of the different categories of anemia in varying proportions. Thus, in the 145 patients with low parasite densities, the anemias: [mild - moderate - no anemia] were respectively [60% (87) - 13% (19) - 27% (39)], for the 34 patients with average parasite density, the proportions were [68% (23) - 6% (2) - 26% (9)], for the 9 patients with high parasite density, the proportions were [44% (4) - 12% (1) - 44% (4)], for the 4 patients with very high parasite density, the values were [50% (2) - 25% (1) - 25% (1)], and for the 2 patients with hyperparasitemia, the proportions were [100% (2)]. Highly affected patients were aged 0-5 years (58), followed by 6-15 years (28) and 16-24 years (27), and were predominantly female (Figure 2).

Clinical manifestations

Malaria attacks were simultaneously associated with several general, neurological, respiratory and/or digestive syndromes. Almost all patients auscultated (98%, 191/194) had general syndromes dominated exclusively by asthenia and/or fever. Just over half the patients (55%, 106/194) had neurological symptoms dominated by headache and/or dizziness (98%, 104/106). Digestive and respiratory syndromes were moderately observed in malaria patients, 48% (95/194) and 31% (61/194), respectively (Figure 3). The main symptoms identified were diarrhea with or without vomiting (89%, 85/95) and cough (82%, 50/61). General, digestive and respiratory syndromes were frequently observed in patients under 16 years of age (0-5 years and 6-15 years). In contrast, neurological and general syndromes were strongly observed in patients aged over 16.

Discussion

The results of this study provide a detailed view of the epidemiological characteristics in the commune of Ntoum, a semi-rural town in the Komo-Mondah department (Gabon) which has experienced a considerable demographic and urban explosion over the last decade.

Prevalence

With prevalences of 13% and 16% [4], Gabon and the Estuary province, respectively, are meso-endemic regions for malaria

[19, 20] The fact that 78% of patients in the study had a positive thick drop reveals that, like most semi-urban towns and villages in Gabon [11- 13, 21], Ntoum is a very active focus of malaria transmission. Indeed, prevalence in Ntoum was on average 3.5 times higher than prevalences reported in other regions of Gabon [9, 10, 22]. As many other studies have noticed, children under 5 years old are the most vulnerable segment of the population and pay the heaviest price for malaria [7, 23-25].,. Among children diagnosed with malaria in Gabon in 2022, 10% lived in urban areas, compared with 35% in rural areas [4]. In addition, our results and those reported in previous studies further highlight the malaria risk factor associated with semi-rural/rural environments [26, 27] and rainy seasons. Indeed, rain falls create a favorable breeding ground for vector proliferation and thus, an increase in mosquito bites at the origin of the peaks of malaria recorded during seasonal transitions between rainy and dry seasons [22, 28]. The predominance of P. falciparum species found in the current study is consistent with its pre- eminent position as a major malaria pathogen in Gabon [22, 29].

Clinical symptoms

The clinical symptoms observed in malaria patients, such as asthenia, fever, headache, dizziness, diarrhea, vomiting and cough, reflect the diversity of malaria manifestations, confirming its polymorphic nature [13, 30]. The fact that 22% (54/248) of patients with a negative thick skin drop display similar syndromes underlines the absolute necessity of implementing and routinely using sensitive in vitro diagnostic tools in departmental health centers in order to make a proven diagnosis of certainty. Indeed, it would seem likely that malaria cases would be carriers of submicroscopic parasitaemia that could not be detected by microscopy but by a high-performance tool such as PCR [21, 31, 32]. The overall malaria prevalence of 78% found in the current study is higher than the 46.2 % reported in 2016 in Dienga (a rural area Southern of Gabon) [21], and the 40% (952/2381) reported in rural, semi-urban, urban cities located in North eastern and South eastern of Gabon [10], and the 6.2 % (269/4342) reported in a large - scale nationwide study [21]. The study size, the sampling period, and the use various diagnostic tools (microscopy, PCR, and RDT) may account for the observed discrepancies among the studies.

Vulnerability and susceptibility to malaria infection

In humans, the intra-erythrocytic life cycle of P. falciparum results in gradual hemolysis of parasitized red blood cells and a fall in hemoglobin levels (anemia) as the infection progresses [16, 33]. In our study, 73% of malaria cases presented with mild or moderate anemia, while 27% of patients were non-anemic. Thus, the vulnerability and susceptibility of individuals to malaria infection would appear to be intrinsically dependent, on the one hand, on genetic factors specific to each patient, as some studies have shown for age and sex, ethnicity, immune system naivety and sickle cell traits associated with reduced parasite blood density [21, 34, 35]. Secondly, the pathogenicity of the infecting parasite, P. falciparum being far more pathogenic than the other human parasites (P. malariae, P. vivax, P. ovale) [28, 30, 36]. With a mean age of 17.83 years, the study population was young and predominantly female (62%, 117/190). Grouping patients by age and sex revealed a marked prevalence among younger patients aged 0-5 years (43%, 81/190), and female patients, which is in line with epidemiological patterns observed in Gabon and other endemic regions [7, 12, 37].

Parasite density and physiological changes

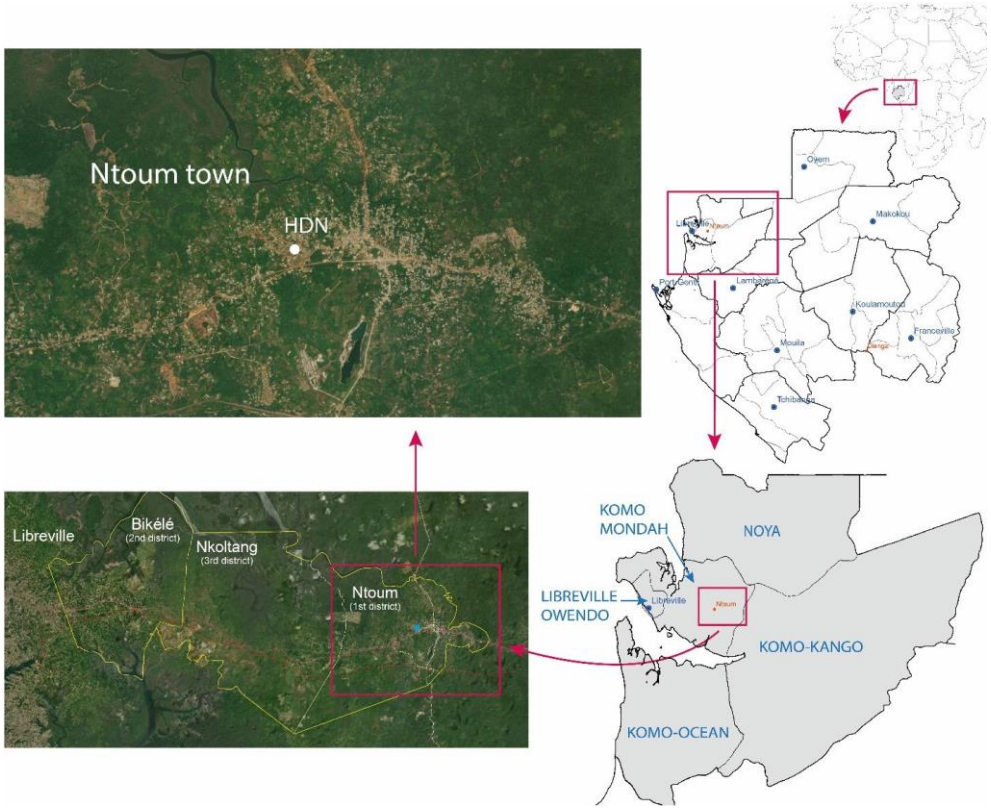
Parasitic infestations in the host caused physiological imbalances, resulting in the simultaneous manifestation of several malarial syndromes (fever, asthenia, headaches, dizziness, dyspnoea). This causality is fairly well described in the literature: when a patient's parasitaemia exceeds a certain threshold, the risk of fever for the individual becomes compelling [38]. Such observations were made in our study, but in view of the parasitaemia (differences) observed, no link could be established between a level of parasitaemia and the development of anaemia in patients, the onset of fever and other syndromes. Patients of the same age/sex with the same parasitaemia manifested anaemia and syndromes of different levels of severity, again pointing to genetic factors in the host [3, 38] and the infecting parasite [28, 30, 36]. As with Mosha *et al*, in our study we observed that parasite density is very high in children and very low in adults [31]. Indeed, the median parasite density in 0-5 year olds (8151 p/µL) was 8 to 4 times higher than that determined in 16-24 year olds (1044p/µL), over 25 year olds (2098 p/µL) and 6-15 year olds (2648 p/µL). Studies of febrile children in four major Gabonese cities between 2005 and 2023 showed fairly similar median parasitemia levels [10, 22].

In endemic areas, immunization increases with age, so the threshold and risk of fever decreases with age [38, 39], as does the onset of syndromes, as observed in our study population. The proportions of patients subject to the various syndromes were much higher in children aged 0-5 years, in contrast to patients aged ≥25 years and 16-24 years, who predominantly manifested general and neurological syndromes. Serious malaria attacks, which are potentially fatal, result from an initial infectious state that remains untreated for a number of days [15, 16, 39] due to negligence or ignorance of the warning symptoms. The fact that no cases of severe malarial anemia were diagnosed underlines the level of awareness of local populations to self-diagnose the pathology. In our study, 51% (99/194) of the anemia encountered were of the hypochromic microcytic type, and in 23 of these cases, the anemia was associated with thrombocytopenia. These results are higher than those observed in India (29%) [40], but lower than those in Senegal (78%) [41]. This proportion of hypochromic microcytic anemia raises concerns about the potential complications of malaria infection.

Conclusion

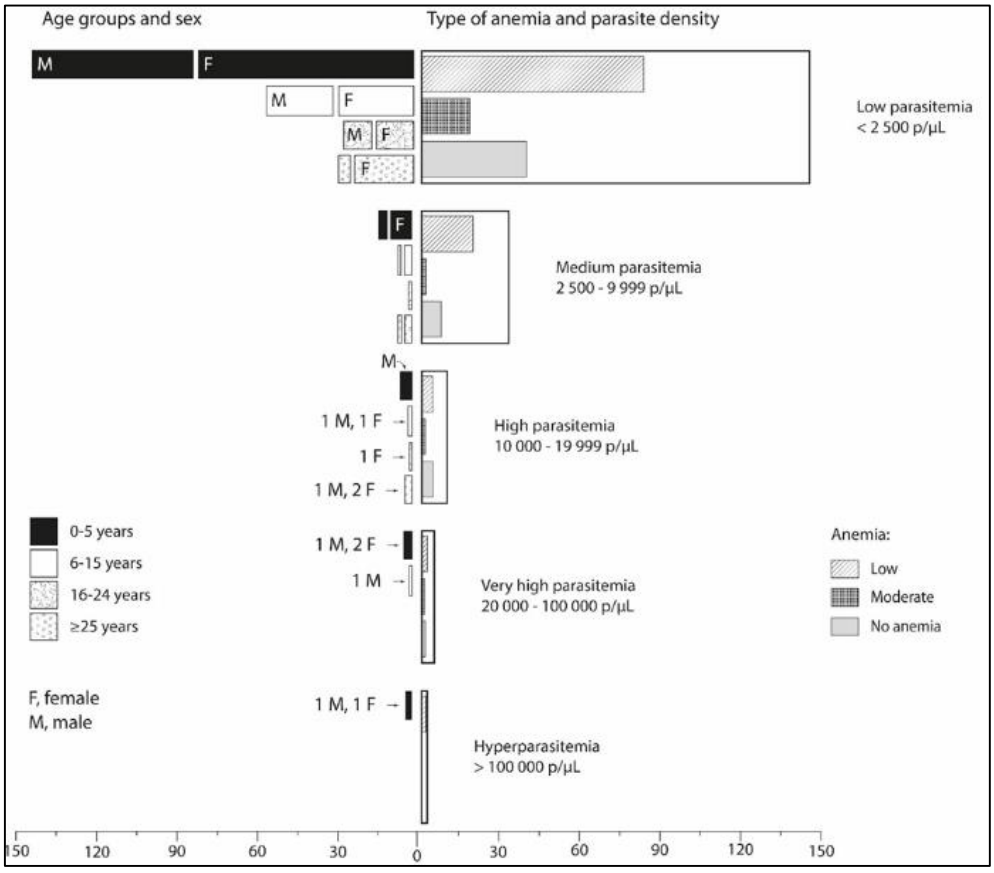
This study has provided crucial information in understanding the occurrence of malaria, and the infectious complications resulting from malaria infection in the Ntoum region. With a prevalence rate of 78%, the results underline the importance and necessity of carrying out targeted, long-term prevention-health education campaigns, and of maintaining epidemiological surveillance in order to effectively combat the disease by reducing the malaria burden.

Looking ahead, we would welcome the opportunity to investigate the proportion and epidemiological impact of patients with submicroscopic parasitaemia in the city as reservoirs of parasite parasites. The same applies to the proportion of parasites and mutant vectors, respectively resistant to the antimalarial molecules and insecticides commonly used in the locality.



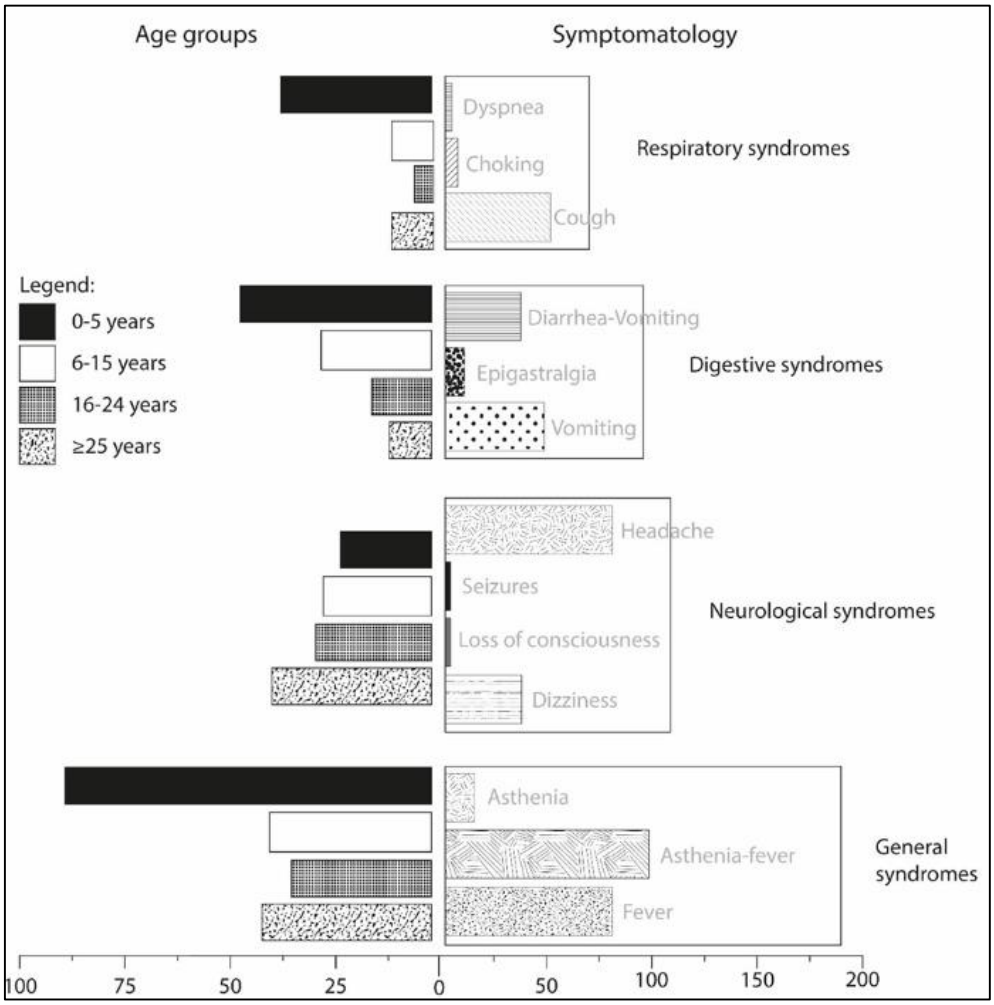
Legend: DNH, Departmental Ntoum Hospital

Fig 1: Geographical location of the study site



Legend: p/µL = parasites/µL blood; F = female; M = male

Fig 2: Diagnosed anemias and observed parasite densities as a function of age



Legend: Tx. = Cough, Diar/vomis. = Diarrhea and vomiting, Vomis. = Vomiting, Cép. = Headache, Asth/fièv = Asthenia and fever

Fig 3: Frequency of clinical signs observed in symptomatic and asymptomatic malaria patients

Table 1: Socio-demographic characteristics of the study population of positive and negative thick drop case

		Frequency (%)		
Variable		**Positive case, *n* = 194 (*78*)**	**Negative case *n* = 54 (*22*)**	**Total, *n* = 248**
Âge (years)	0-5	83 (*43*)	15 (*26*)	98 (*39*)
	6-15	37 (*19*)	6 (*10*)	43 (*17*)
	16-24	33 (*17*)	14 (*24*)	47 (*19*)
	≥25	41 (*21*)	23 (*40*)	64 (*25*)
Gender	Male	74 (*38*)	23(*40*)	97 (*39*)
	Female	120 (*62*)	35 (*60*)	155 (*61*)
Educational background	Primary	16 (*8*)	7 (*12*)	23 (*9*)
	Secondary	136 (*70*)	36 (*62*)	172 (*68*)
	Universsity	42 (*22*)	15 (*26*)	57 (*23*)
Occupation	none	66 (*34*)	16 (*27*)	82 (*33*)
	Pupil/Student	64 (*33*)	16 (*27*)	80 (*32*)
	Self-employed	27 (*14*)	9 (*16*)	36 (*15*)
	Civil servant	25 (*13*)	11 (*19*)	36 (*14*)
	Employee	5 (*2*)	4 (*7*)	9 (*3*)
	Farmer	5 (*3*)		5 (2)
	Administrative	1 (*0,5*)	1 (*2*)	2 (1)
	Religious		1 (*2*)	1 (*0,5*)
	Sportsman	1 (*0,5*)		1 (*0,5*)
Town of residence	Ntoum	181 (*93*)	55 (*95*)	236 (*93*)
	Kango	7 (*4*)	2 (*3*)	9 (*4*)
	Libreville	6 (*3*)	1 (*2*)	7 (*3*)

Table 2: Blood count profiles by age of patients, n = 194

Variables	Total	Class of age (years) Frequency (%)				Thrombocyte	
		0-5	6-15	16-24	≥ 25	thrombocytopenia	thrombocytosis
malarial infections with anemia, Hb < 11 g/dL	**140 (73)**	**70**	**26**	**17**	**23**	**33**	**34**
Moderate anemia, 5 ≥ Hb < 8 g/dL	23 (17)	18	1	2	2	7	4
Hypochromic normocytosis	17 (74)	15	1		1	6	2
Normocytic microcytosis	3 (13)	2			1	1	1
Hypochromic normocytosis	2 (9)	1			1		1
Normochromic normocytosis	1 (4)			1			
Low anemia, 8 ≥ Hb < 11 g/dL	118 (83)	52	25	16	21	26	30
Hypochromic microcytosis	82 (71)	42	21	10	9	17	24
Normocytic microcytosis	8 (7)	5	1		2	2	1
Hypochromic normocytosis	15 (13)	3	2	4	6	3	5
Normochromic normocytosis	12 (8)	4	1	2	5	4	2
Normochromic macrocytosis	1 (1)			1		1	
malarial infections without anemia, Hb > 11 g/dL	**53 (27)**	**11**	**12**	**14**	**16**	**14**	**15**
Hypochromic microcytosis	13 (24)	5	3	3	2	1	5
Normocytic microcytosis	1 (2)			1			
Hypochromic/ Normochromic normocytosis	37 (70)	6	9	10	12	13	8
Normochromic macrocytosis	2 (4)				2		2

References

1. Lalremruata A, Jeyaraj S, Engleitner T, Joanny F, Lang A, Bélard S, Mombo-Ngoma G, Ramharter M, Kremsner PG, Mordmüller B, Held J: Species and genotype diversity of Plasmodium in malaria patients from Gabon analysed by next generation sequencing. Malar J. 2017; 16:398.
2. World Health Organization: World malaria report, 2023.
3. World Health Organization: World malaria report, 2022.
4. Direction Générale de la Statistique (DGS): Enquête Démographique et de Santé au Gabon, 2019-21. 2023; 1:691.
5. Direction Générale de la Statistique (DGS): Recensement général de la population et des logements de 2013 du Gabon. RGPL, 2013.
6. Lendongo-Wombo J-B, Oyegue- Liabagui S-L, Biteghe-Bi-Essone J-C, Ngoungou EB, Lekana-Douki J-B: Epidémiology of malaria from 2019 to 2021 in the southeastern city of Franceville, Gabon. BMC Public Health. 2022; 22:2313.
7. Boundenga L, Bignoumba M, Dibakou SE, Mombo LE, Moukagni- Mussadji CJ, Wora DM, Kassa-Kassa F, Bisseye C, Onanga R: Decrease on malaria clinical cases from 2017 to 2019 in Franceville, Southeast Gabon, Central Africa. J Public Health Afr. 2023; 14:1865.
8. Mourou JR, Coffinet T, Jarjaval F, Cotteaux C, Pradines E, Godefroy L, *et al*. Malaria transmission in Libreville: results of a one year survey. Malar J. 2012; 11:40.
9. Pamba R, Koumba AA, Zinga-Koumba CR, Kutomy POO, Ovono APM, Itsiembou LD, *et al*. Six years surveillance of Plasmodium falciparum malaria among febrile patients reporting in Centre de Santé d'Okala, North West Gabon. Microbes and Infectious Diseases. 2021; 2:823-830.
10. Kouna LC, Oyegue-Liabagui SL, Voumbo-Matoumona DF, Lekana-Douki JB. Malaria Prevalence in Asymptomatic and Symptomatic Children Living in Rural, Semi-Urban and Urban Areas in Eastern Gabon. Acta Parasitologica, 2024, 1- 12.
11. Bisseye C, Kombila A, Ndong Mba T, Ngossanga B, Moundounga Kenguele H. Prevalence of Malaria from Blood Smears: A Four-Year Retrospective Study from the Jean Claude Handrault Hospital, Southern Gabon. International Journal of Tropical Disease & Health. 2021; 42:1-7.
12. Imboumy-Limoukou RK, Lendongo-Wombo JB, Nguimbyangue-Apangome AF, Biteghe Bi Essone J-C, Mounioko F, Oyegue-Libagui LS, *et al*. Severe malaria in Gabon: epidemiological, clinical and laboratory features in Amissa Bongo Hospital of Franceville. Malaria Journal. 2023; 22:1-8.
13. Lendzele SS, Rodriquge MN, Boris M, Kutomy PO, Armel KA, Christophe Z-KR, Leotard SO, Mavoungou JF: Is malaria cases frequency correlated with the environmental and demographic composition at Mitzic medical center in Gabon? DYSONA-Life Science 2022, 3:41-48.
14. Ebigre JM, Et Marius C. Etude d'une séquence mangrove-tanne en milieu équatorial, baie de la mondah (GABON). Travaux et Documents de Géographie Tropicale CEGET, 1984. 2ème trimestre, n° 51
15. Organization WH: Guidelines for the treatment of malaria. World Health Organization, 2015.
16. White NJ. Anaemia and malaria. Malar J. 2018; 17:371.
17. Nkhoma P, Loti P, Sinkala M, Mantina H, Mwaba F, Kafita D, *et al*. Anaemia Types and Severity in Patients aged 1 to 14 years at the Children's Hospital of the University Teaching Hospitals in Zambia. Med Rxiv 2020:2020.2009. 2017.20194779.
18. Planche T, Krishna S, Kombila M, Engel K, Faucher J, Ngou-Milama E, *et al*. Comparison of methods for the rapid laboratory assessment of children with malaria. The American journal of tropical medicine and hygiene. 2001; 65:599-602.
19. Boudin C, Robert V, Gazin P. Évaluation épidémiologique des paludismes.
20. Organization WH. Report on the Malaria Conference in Equatorial Africa, held under the joint auspices of the World Health Organization and the Commission for Technical Co- operation in Africa South of the Sahara, Kampala, Uganda, 27 November-9 December 1950. World Health Organization; 1951.
21. Pegha Moukandja I, Biteghe Bi Essone JC, Sagara I, Kassa Kassa RF, Ondzaga J, Lékana Douki JB, Bouyou

Akotet M, Nkoghe Mba D, Touré Ndouo FS. Marked Rise in the Prevalence of Asymptomatic Plasmodium falciparum Infection in Rural Gabon. PLoS One. 2016; 11:e0153899.
22. Maghendji-Nzondo S, Nzoughe H, Lemamy GJ, Kouna LC, Pegha- Moukandja I, Lekoulou F, Mbatchi B, Toure-Ndouo F, Lekana-Douki JB. Prevalence of malaria, prevention measures, and main clinical features in febrile children admitted to the Franceville Regional Hospital, Gabon. Parasite. 2016; 23.
23. Ajakaye OG, Ibukunoluwa MR. Prevalence and risk of malaria, anemia and malnutrition among children in IDPs camp in Edo State, Nigeria. Parasite Epidemiol Control. 2020; 8:e00127.
24. Makenga G, Menon S, Baraka V, Minja DT, Nakato S, Delgado-Ratto C, Francis F, Lusingu JP, Van Geertruyden JP. Prevalence of malaria parasitaemia in school- aged children and pregnant women in endemic settings of sub-Saharan Africa: a systematic review and meta-analysis. Parasite Epidemiol Control. 2020; 11:e00188.
25. Tsegaye AT, Ayele A, Birhanu S. Prevalence and associated factors of malaria in children under the age of five years in Wogera district, northwest Ethiopia: A cross-sectional study. PLoS One. 2021; 16:e0257944.
26. Duguay C, Mosha JF, Lukole E, Mangalu D, Thickstun C, Mallya E, Aziz T, Feng C, Protopopoff N, Mosha F. Assessing risk factors for malaria and schistosomiasis among children in Misungwi, Tanzania, an area of co-endemicity: A mixed methods study. PLOS Global Public Health. 2023; 3:e0002468.
27. Chilot D, Mondelaers A, Alem AZ, Asres MS, Yimer MA, Toni AT, Ayele TA. Pooled prevalence and risk factors of malaria among children aged 6–59 months in 13 sub-Saharan African countries: A multilevel analysis using recent malaria indicator surveys. PLoS One. 2023; 18:e0285265.
28. Drakeley C, Abdulla S, Agnandji ST, Fernandes JF, Kremsner P, Lell B, Mewono L, Bache BE, Mihayo MG, Juma O, *et al*. Longitudinal estimation of Plasmodium falciparum prevalence in relation to malaria prevention measures in six sub-Saharan African countries. Malar J. 2017; 16:433.
29. Zoleko Manego R, Koehne E, Kreidenweiss A, Nzigou Mombo B, Adegbite BR, Dimessa Mbadinga LB, Akinosho M, Matthewman J, Adegnika AA, Ramharter M, Mombo-Ngoma G. Description of Plasmodium falciparum infections in central Gabon demonstrating high parasite densities among symptomatic adolescents and adults. Malar J. 2019; 18:371.
30. Nateghpour M, Hosseininasab A, Farrokhnia M, Dastouri F, Alidoosti K, Sadequi D, Ahmadi A. Species-dependent clinical findings of malaria caused by various plasmodia in an endemic area of Kerman Province, Southeastern Iran. Iranian J Public Health. 2017; 46:525.
31. Mosha JF, Sturrock HJW, Greenhouse B, Greenwood B, Sutherland CJ, Gadalla N, Atwal S, Drakeley C, Kibiki G, Bousema T, *et al*. Epidemiology of subpatent Plasmodium falciparum infection: implications for detection of hotspots with imperfect diagnostics. Malar J. 2013; 12:221.
32. Okell LC, Bousema T, Griffin JT, Ouédraogo AL, Ghani AC, Drakeley CJ. Factors determining the occurrence of submicroscopic malaria infections and their relevance for control. Nat Commun. 2012; 3:1237.
33. Bashawri LA, Mandil AA, Bahnassy AA, Ahmed MA. Malaria: hematological aspects. Ann Saudi Med. 2002; 22:372-376.
34. Cao H, Vickers MA. Oxidative Bashawri LA, Mandil AA, Bahnassy AA, Ahmed MA. Malaria: hematological aspects. Ann Saudi Med. 2002; 22:372-376.
35. Chao DL, Oron AP, Chabot-Couture G, Sopekan A, Nnebe-Agumadu U, Bates I, Piel FB, Nnodu O. Contribution of malaria and sickle cell disease to anaemia among children aged 6–59 months in Nigeria: a cross-sectional study using data from the 2018 Demographic and Health Survey. BMJ Open. 2022; 12:e063369.
36. Kotepui M, Piwkham D, PhunPhuech B, Phiwklam N, Chupeerach C, Duangmano S. Effects of malaria parasite density on blood cell parameters. PLoS One. 2015; 10:e0121057.
37. Nzabakiriraho JD, Gayawan E. Geostatistical modeling of malaria prevalence among under-five children in Rwanda. BMC Public Health. 2021; 21:1-11.
38. Rogier C, Commenges D, Trape JF. Evidence for an age-dependent pyrogenic threshold of Plasmodium falciparum parasitemia in highly endemic populations. Am J Trop Med Hyg. 1996; 54:613-619.
39. Newton CR, Krishna S. Severe falciparum malaria in children: current understanding of pathophysiology and supportive treatment. Pharmacol Ther. 1998; 79:1-53.
40. Agarwal P. Haematological Parameters in Malaria: A Clinico Patho
41. Diagne I, Fall A, Diagne-Gueye N, Déme-Ly I, Lopez-Sall P, Faye C, Sarr M, Camara B, Sow H. Hypochromic microcytic anaemia in pediatrics: Frequency and response to iron treatment. A study in outpatients in Albert Royer National Children Hospital of Dakar, Senegal. J Pediatr Pueric. 2010; 23:119-124