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Lipid Profile Alterations (*dyslipidemia*) in Adults during *Plasmodium falciparum* Infection in the City of Butembo, Northeastern Democratic Republic of Congo

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Abstract: *Malaria remains a major public health concern in sub-Saharan Africa, not only for its high morbidity and for mortality rates but for the complexity, it introduces in clinical diagnosis, especially in resource-limited settings. In this context, exploring alternative biological markers that could support or refine diagnosis becomes crucial. In this cross-sectional study conducted at the Internal Medicine and Parasitology Department of Matanda Hospital in Butembo, a distinct pattern of lipid alterations was identified among adult patients with confirmed* Plasmodium falciparum *malaria. Spanning from July 24, 2023, to November 12, 2024, the investigation compared 245 febrile patients with positive thick and thin blood films to 253 febrile controls with negative parasitological results. The findings revealed a consistent profile of dyslipidemia in malaria cases, notably marked by hypocholesterolemia, reduced HDL cholesterol levels, elevated triglycerides, and a general decline in total cholesterol. These lipid disruptions were significantly associated with higher levels of parasitaemia, suggesting a potential diagnostic utility. Moreover, the study highlighted that such lipid anomalies could serve as valuable early indicators for malaria diagnosis, particularly in settings where parasitological confirmation may be delayed or inconclusive. This underscores the clinical relevance of lipid profiling in the prompt recognition and management of malaria, ultimately contributing to the prevention of severe and potentially fatal outcomes.*

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Keywords: malaria, plasmodium falciparum, dyslipidemia, lipid profile, parasitic infection, adults, butembo, Democratic Republic of Congo (DRC)

Résumé : Le paludisme demeure un problème majeur de santé publique en Afrique subsaharienne, tant par sa forte morbidité que par la complexité de son diagnostic, notamment dans les milieux à ressources limitées. Dans ce contexte, l'exploration de marqueurs biologiques alternatifs susceptibles de soutenir ou de compléter le diagnostic s'avère d'une importance capitale. La présente étude transversale, menée au sein du service de Médecine interne et de Parasitologie de l'Hôpital Matanda à Butembo, met en évidence un profil lipidique particulier chez les adultes atteints de paludisme à *Plasmodium falciparum*. Réalisée du 24 juillet 2023 au 12 novembre 2024, elle a porté sur 245 patients fébriles présentant des frottis sanguins (goutte épaisse et lame mince) positifs, comparés à un groupe contrôle de 253 patients fébriles aux frottis négatifs. Les résultats révèlent une dyslipidémie constante chez les sujets infectés, caractérisée par une hypocholestérolémie, une hypoHDLémie, une baisse du cholestérol total, ainsi qu'une hypertriglycéridémie. Ces altérations lipidiques se sont avérées significativement corrélées à un taux élevé de parasitémie, suggérant leur utilité potentielle à des fins diagnostiques. Par ailleurs, l'étude souligne que de telles anomalies lipidiques pourraient constituer des indices précoces pertinents permettant d'orienter le diagnostic du paludisme, en particulier dans les situations où les résultats parasitologiques sont absents, retardés ou incertains. Ainsi, le profil lipidique pourrait contribuer à une prise en charge rapide et efficace de la maladie, prévenant l'apparition de complications graves, parfois mortelles.

Mots-clés : Paludisme, Plasmodium falciparum, Dyslipidémie, Profil lipidique, Adultes, Infection parasitaire, Butembo, République Démocratique du Congo (RDC), etc.

INTRODUCTION

Malaria remains a major public health concern, with approximately 14 million new cases reported in 2020 alone [1]. Its burden is disproportionately high in low- and middle-income countries, particularly in those situated within the WHO African Region [2]. Recent estimates attribute over 69,000 malaria-related deaths to the disruption of prevention, diagnostic, and treatment services [2], including insufficient attention to early detection of metabolic changes during infection [3]. In endemic regions such as tropical and subtropical areas of Africa, Southeast Asia, Latin America, and the Middle East, severe malaria is responsible for 11 to 30% of deaths [4,5], with *Plasmodium falciparum* accounting for 99.7% of all cases [2,6].

Infection with *P. falciparum* is frequently associated with multi-organ failure, often accompanied by a constellation of clinical signs, hematological anomalies, and biochemical disturbances that necessitate hospitalization and targeted clinical intervention [7]. Among these disturbances, significant alterations in serum lipid profiles have been consistently observed during the course of malaria. A 2013 systematic review and meta-analysis reported notable reductions in total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), and low-density lipoprotein cholesterol (LDL-C) among malaria patients compared to healthy individuals [8]. While not specific, these lipid abnormalities may serve as useful biological indicators in febrile patients from or returning from endemic areas [9,10].

Although microscopic examination of Giemsa-stained thin and thick blood smears remains the diagnostic gold standard [11,12], its sensitivity may be limited, particularly when parasitemia is low or when the smear quality is suboptimal [10]. Diagnostic accuracy is further influenced by variables such as the examiner's expertise, endemicity level, host immunity, underlying hemoglobinopathies, genetic background, and the specific parasite strain [10].

Malaria is known to induce a wide range of hematological and biochemical disturbances, including anemia, lymphopenia, thrombocytopenia, marked hypocholesterolemia, hypoHDLemia, hypertriglyceridemia, and elevated levels of CRP and LDH [1,7,11,14]. Accordingly, the use of biochemical or hematological markers is increasingly being explored as a complementary tool in the management of malaria [11]. Other studies have also reported fibrinogen abnormalities and prolonged coagulation times, including prothrombin time [15].

Importantly, emerging evidence indicates that these metabolic alterations are significantly correlated with parasitemia levels [37]. However, their specific burden remains poorly characterized among patients with malaria attending primary healthcare facilities in endemic regions such as the eastern Democratic Republic of Congo (DRC). This region, known for its mountainous geography and unstable malaria transmission patterns, exhibits a malaria prevalence of approximately 3% [16]. Transmission is highly seasonal, marked by fewer than two infective mosquito bites per person annually, resulting in low levels of acquired immunity and rendering the population vulnerable to periodic malaria epidemics [17].

Despite the DRC being the second most malaria-affected country globally, there remains a paucity of data on the hematological and biochemical implications of malaria within its borders. This study, therefore, aims to characterize the lipid profile alterations associated with *P. falciparum* infection among adult patients seeking care in an unstable malaria transmission zone in the eastern DRC.

METHODS

Study design and setting

We conducted this analytical cross-sectional study among patients attending the Internal Medicine and Parasitology Departments of Matanda Hospital, situated in the city of Butembo, in the North Kivu province of the Democratic Republic of Congo, between July 24, 2023, and November 12, 2024. Matanda Hospital was purposefully selected as the study site due to its central role in the management of diverse medical conditions, notably malaria, diabetes mellitus, and hypercholesterolemia. As one of the few healthcare institutions in Butembo staffed with trained medical specialists, Matanda holds a prominent position within the region. Nestled in a mountainous landscape at altitudes ranging from 1,000 to 2,000 meters, Butembo is a densely populated urban center, home to nearly four million inhabitants. Matanda Hospital also functions as a teaching and tertiary referral center, providing care and clinical training across the health zones of Katwa and Butembo.

Study population

Participants in this study were selected from individuals who sought medical attention at Matanda Hospital during the designated study period. Febrile adult patients presenting to the Internal Medicine and Parasitology departments were purposively enrolled. Two distinct subgroups were constituted: the first comprised patients exhibiting fever and confirmed positive for malaria through thick and thin blood smear microscopy; the second, serving as the control group, included febrile patients who tested negative on the same diagnostic test. The minimum required sample size was calculated using G*Power software version 3.1.9.4 [18–19]. 498 participants were included in the final analysis 245 patients diagnosed with malaria and 253 classified as malaria-negative febrile individuals.

Study procedures

Four independent research assistants carried out purposive data collection using a semi-structured questionnaire designed to capture both socio-demographic characteristics and biological parameters, notably the lipid profile (total cholesterol, HDL cholesterol, LDL cholesterol, and triglycerides), as well as malaria parasitaemia. Individuals expressing willingness to participate were approached, provided with detailed information about the study, and invited to complete the survey upon giving their informed consent. A multi-stage purposive sampling strategy was employed. Initially, demographic and clinical data were collected using a structured extraction form. Subsequently, each participant was referred to the laboratory for comprehensive biological and parasitological analyses.

Laboratory analysis

The diagnosis of malaria was biologically confirmed with thick blood films and peripheral blood smears. Three qualified laboratory technicians determined parasite density independently. The thick film was stained using 10% Giemsa for 10 minutes, while the blood smear was processed with May-Grünwald Giemsa stain. Each technician examined the slides independently under a 100X oil immersion objective. For every confirmed case, the asexual forms of *Plasmodium* parasites were counted against 200 leukocytes. The final parasitaemia level was derived as the mean of the three separate counts. Parasite density was calculated using the following formula:

Parasites/µL = (Parasites counted / 200) × Total WBC count [21].

Based on this quantification, parasitaemia was categorized into three levels:

- Low parasitemia: < 1,000 parasites/µL
- Moderate parasitemia: 1,000–4,999 parasites/µL
- High parasitemia: ≥ 5,000 parasites/µL [19].

Operationally, a malaria case was defined as a febrile patient with a positive result from either the thick blood film or the blood smear showing the presence of *Plasmodium* parasites.

The lipid profile in malaria-positive patients was analyzed through standardized biochemical procedures. For each selected participant, 3 mL of venous blood was collected into tubes containing ethylene-diamine-tetra-acetic acid (EDTA) to prevent coagulation, enabling a complete blood count and lipid analysis. Biochemical measurements were performed using an automated photometric analyzer. Dyslipidemia was defined according to the National Cholesterol Education Program Adult Treatment Panel III (NCEP/ATP III) guidelines, as the presence of one or more of the following lipid abnormalities:

- Total cholesterol (TC) ≥ 200 mg/dL
- Triglycerides (TG) ≥ 150 mg/dL
- Low-density lipoprotein (LDL) ≥ 160 mg/dL
- High-density lipoprotein (HDL) < 40 mg/dL [20].

To assess the diagnostic relevance of lipid alterations, the sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) of each lipid parameter were computed using GMRC software version 3. Concordance between lipid abnormalities and malaria diagnosis was assessed using Cohen's Kappa coefficient, interpreted as follows:

Very weak agreement: Kappa = 0–0.20
(ii) Weak agreement: Kappa = 0.21–0.40
(iii) Moderate agreement: Kappa = 0.41–0.60
(iv) Strong agreement: Kappa = 0.61–0.80
(v) Almost perfect agreement: Kappa = 0.81–1.00 [16–18].

Statistical analysis

Data were initially entered into Microsoft Excel and subsequently exported to SPSS for comprehensive statistical analysis. Descriptive statistics were presented as frequencies and percentages for categorical variables, while means and medians were calculated for continuous variables. Inferential analyses were conducted using Student's t-test, Fisher's exact test, and the Chi-square test to identify factors associated with lipid profile alterations in malaria-infected patients. A p-value of less than 0.05 was considered statistically significant. Associations were further evaluated through the calculation of odds ratios.

Ethical considerations

The Ethics Committee of the University of Kisangani approved all procedures undertaken in this study. Formal authorization to conduct the research was granted by the Head of Department at the University of Kisangani Teaching Hospital, the Executive Director of Matanda Hospital, and the Coordinator of the Katwa Health Zone. The study was carried out in full compliance with the ethical principles outlined in the Declaration of Helsinki. Written informed consent was obtained from all participants prior to their inclusion in the study.

RESULTS

General characteristics of the study population.

Between July 24, 2023 and November 12, 2024, 498 febrile patients sought medical attention at the Internal Medicine Department of Matanda Hospital. Among them, 245 individuals (49.2%) tested positive for malaria, while 253 (50.8%) tested negative, serving as the control group. The majority of malaria-positive patients were aged between 21 and 40 years, and notably, over half (216 individuals, or 88.2%) presented with a high level of parasitaemia. Biochemical analysis revealed that most malaria-infected patients exhibited significant lipid disturbances: 79.6% had reduced total cholesterol, 99.6% presented with low HDL cholesterol levels, and 73.1% showed elevated triglyceride concentrations.

Statistical analysis demonstrated no significant association between malaria infection and demographic variables such as age, gender, or occupation ($p > 0.05$). However, a significant

difference was observed in lipid profiles: the mean total cholesterol level among malaria patients was markedly lower (117.7 ± 48.7 mg/dL) compared to the control group (168 ± 29.3 mg/dL). Hypocholesterolemia was found in 79.6% of malaria cases versus 44.7% in controls, while low HDL cholesterol was observed in 73.1% of infected individuals compared to 28.9% in non-infected counterparts (Table 1).

Table 1: Baseline information of study participants

Variables	Cases (N=245)	Control (N=253)	Chi-square	p-value
Socio-demographic factors				
Age in years (mean±SD)	35 ± 8.3	39±7.9	8.90	>0.05
Age group			11.53	<0.05
>40 years	43 (17.6%)	121 (47.8%)		
21-40 years	146 (59.6%)	121 (47.8%)		
15-20 years	56 (22.9%)	11 (4.3%)		
Gender			3.25	>0.05
Male	112 (45.7%)	137 (54.2%)		
Female	133 (54.3%)	116 (45.8%)		
Occupation			10.8	>0.05
Farmer	145 (60%)	159 (62.8%)		
Business worker	37 (16%)	35 (13.8%)		
Students	31 (13%)	40 (15.8%)		
Teachers	17 (6.9%)	11 (4.3%)		
Others	15 (6.1%)	8 (3.2%)		
Malaria parasite density				
Mean parasite density (Mean±SD)	21575.6 ±13407.1			
High density	216 (88.2%)			
Moderate density	20 (8.2%)			
Low density	9 (3.6%)			
Total cholesterol (Mean±SD)	117 ± 48.7	168 ± 29.3	5.47	<0.05
<200	195 (79.6%)	113 (44.7%)		
200-240	50 (20.4%)	140 (55.3%)		
HDL cholesterol			7.54	<0.05
Low < 40 mg/dl	244 (99.6%)	93 (36.8%)		
Normal ≥ 40 mg/dl	1 (0.4%)	160 (63.2%)		
Triglycerides			2.85	<0.05
<150	66 (26.9%)	180 (71.1%)		
≥150	179 (73.1%)	73 (28.9%)		

Lipid profile in relation to parasitemia patients with malaria

We found a statistically significant difference between the parasite level and the lipid abnormalities during malaria infection.

Table 2: Association between parasitemia and lipid changes in cases with malaria.

Parasite density				
Variables	Low (N=9)	Moderate (N=20)	High (N=216)	p-value/Fisher
Total cholesterol				
<200	5 (55.6%)	11 (55%)	179 (82.9%)	<0.05
200-240	4 (44.4%)	9 (45%)	39 (17.1%)	
HDL cholesterol				
Low<40 mg/dl	9 (100%)	20 (100%)	215 (99.5%)	<0.05
Normal ≥40 mg/dl	0 (0%)	0 (0%)	1 (0.5%)	
Triglycerides				
< 150	4 (44.4%)	11(55%)	51(23.3%)	<0.05
≥ 150	5 (55.6%)	9 (45%)	165(76.4%)	

Sensitivity, specificity, positive predictive value, and negative predictive value of lipid parameters.

We notice that hypoHDLemia, hypertriglyceridemia, lower total cholesterol are reliable in predicting malaria.

Table 3: Specificity, sensitivity, positive predictive value (PPV), negative predictive value (NPV) and the Kappa coefficient of lipid parameters.

Variable	Sensitivity	Specificity	VPP	VPN	KAPPA (%)
HypoHDLemia	0,41	36,76	0,62	27,6	63
Low Cholesterol total	20,41	44,66	26,32	36,69	35
Hypertriglyceridemia	73,06	71,15	71,15	73,17	44

DISCUSSION

Socio-demographic characteristics.

This study revealed that most of our patients were of female sex, of profession respectively farmer and trader. The age group from 21 to 40 years was the more represented and the sex ratio of 1.17 in favor of female malaria patients (table 1). This sex ratio value is slightly lower than that found

by Nlinwe and Nange in their study where the sex ratio value found was 1.38 in favor of female [22]. In Butembo, agriculture is generally a female activity, the same is true of commercial activity; the 21 to 40 age group is the most productive age group of the population. The search for fertile land leads to the displacement of the population, often to endemic malarial areas.

Lipid profile: Cholesterolemia, HDL cholesterol and triglycerides.

Hypocholesterolemia was found in 79.6% of cases (Table 1). Our results are similar to those found by Chukuoka et al in Nigeria who found that 84% of malaria patients had hypocholesterolemia [23]. The value found during our study is clearly higher than those of the series of Badiaga et al. [24] and Chagnon et al. [25] in France, which were respectively 40% and 45%. In our study, the decrease in total cholesterol and HDL cholesterol was observed in patients with malaria compared to those in the control group. Lowering total cholesterol and HDL cholesterol has been documented for a long time. The links between infection with fever and hypocholesterolemia have been identified for a long time. In 1960, Justin-Besançon et al. specified that two out of three bacterial diseases positively modify cholesterolaemia [26]. Similarly, many authors have demonstrated a change in lipid levels during a febrile or septic state. The association of hypocholesterolemia and hypertriglyceridemia is more frequently found during the acute phase of an infection. These changes are accentuated in the first three days of infection [27].

HypoHDLemia, observed in 99.6% of cases (Table1), can be explained by an inhibition, by the parasite or its products, of lecithin-cholesterol acyl transferase and lipoprotein lipase, which participate in the development of HDLc [29]. Our results are similar to those obtained in the series of Baptisa JL et al. [30] and Ozkaya G et al. [31], for whom, patients infected with P. falciparum had a significantly lower cholesterol level compared to the group of healthy and normal controls. They also found an inverse correlation between parasitaemia and cholesterol levels. The latter was considered a specific indicator (98%) of Plasmodium infection [24].

Malaria is involved in the regulation of cholesterol. Hypocholesterolemia and hypertriglyceridemia have been observed in uncomplicated and complicated malaria. The membrane cholesterol plays a role in the pathogenesis, immune evasion and clinical manifestations of P. falciparum malaria. A study conducted in Saudi Arabia showed that there is a significant inverse correlation between parasitaemia and cholesterol levels in patients. The erythrocytic stages of Plasmodium have no reserve lipids and are unable to synthesize fatty acids or cholesterol themselves de novo. They are forced to incorporate lipids from their environment into the host to satisfy the lipoprotein requirements of schizogony [32]. Thus, we can explain the differences in lipid levels during malaria by the degree of parasitaemia, itself depending on many factors such as

the epidemic facies, the number of bites, the plasmodial species, the environment, the use of insecticide-treated mosquito nets.

Hypertriglyceridemia was found in 73.1% of malaria cases. Hypertriglyceridemia can be explained by the reduction in the activity of lipoprotein lipase, in particular under the influence of tumor necrosis factor (TNF), and by a deficiency in the purification of triglycerides from the plasma compartment [30]. This hypertriglyceridemia has long been linked to the risk of cardiovascular disease, myocardial infarction, coronary artery disease and death in adults, independently of the other components of the lipid profile, hence the problem of this finding [35,36]. Ultimately, it seems that malaria has the singularity of lowering, in a non-negligible number of cases, cholesterolemia more than most other febrile infectious states, which could give such a finding additional diagnostic value. Our results corroborate those found by Visser et al who had found, during their studies, that triglycerides were elevated in febrile patients suffering from malaria compared to the control group [37]. The hypothesis is based on the tumor necrosis factor's ability to inhibit lipoprotein lipase, which is increased during malaria attacks [38].

Our results agree with those obtained by Ozkaya G et al who had observed low cholesterol levels and high triglyceride levels in patients infected with malaria compared to normal healthy controls [39]. Similarly, Al Omar IA et al reported changes in plasma lipoproteins in acute malaria resulting from a decrease in HDLc and LDLc levels and a moderate increase in triglycerides [40, 41]. In this regard, other investigators have found significant changes in the lipid profile in malaria-endemic areas in subjects infected with P. falciparum [42].

In a systematic review and meta-analysis performed by Visser et al [37] to determine a lipid profile of changes in malaria, the following conclusions were drawn: concentrations of total cholesterol, high-density lipoprotein (HDL) and low-density lipoproteins (LDL), were lower in malaria and other febrile illnesses compared to healthy controls. The decline was more marked and statistically significant in malaria compared to other febrile illnesses. These results were consistent across the included studies. Triglycerides were higher compared to healthy controls, but not statistically significant compared to symptomatic controls. In conclusion, this meta-analysis suggested that the observed changes in lipid profiles are considered characteristic for malaria [37]. Plausible hypotheses of biological mechanisms involving host lipid changes and the pathogenesis of malaria exist.

The changes in the lipid profile including total hypocholesterolemia, hypoHDLemia and hypertriglyceridemia are more marked in case of high parasitemia with a significant difference [23, 41,37]. This corroborates the results of our research.

In our study, the sensitivity, specificity, positive predictive value and negative predictive value of biological parameters were determined (table3). Among these parameters, hypoHDLemia, hypertriglyceridemia, lower total cholesterol were reliable in predicting malaria (Table 3). The coefficient of concordance of the results to predict malaria in adults in Butembo was strong in the case of hypoHDLemia; moderate in case of hypertriglyceridaemia and low in case of lower total cholesterol. Our results are similar to those found by other authors, including Lithia. B et al [45], Chukwoka UM et al [23], Kotepui M et al [43] and Gebreweld A et al [44], who proved, during their research, that lower total cholesterol and HDL cholesterol, hypertriglyceridaemia were predictive factors of malaria in adult subjects, given their sensitivities and specificities.

Limitations of the study and outlook

This study provided important results on adult malaria in Butembo. However, it has some limitations. The small sample size may have underestimated or overestimated the prevalence of the parameters studied. The convenience sample was not representative of the city's adult population. Nevertheless, our study made it possible to generate hypotheses, which constituted the bulk of our conclusions. Other comparative studies are necessary to confirm or refute them.

CONCLUSION

Notable variations of some biological parameters were recorded during the bout of Plasmodium falciparum malaria in adults in the city of BUTEMBO. These parameters could be useful in the diagnosis of malaria and/or be an indicator of the severity of the disease. Lowered total cholesterol and hypoHDLemia, hypertriglyceridemia have been constant disturbances in adult malaria cases in the city of BUTEMBO. These lipid abnormalities are significantly associated with elevated parasitaemia.

- **Authors' Contributions**

KMJB conceived and designed the study MBV collected, analyzed, and drafted the manuscript. KTC supervised the research process for this study BAS reviewed the manuscript for the accuracy of its technical and intellectual content. All authors approved the final version of the manuscript.

- **Role of funding source**

This study did not receive any funding. The authors provided the financial support and it had no role in study design, data collection, analysis, or interpretation of data as well as the submission for publication.

- **Availability of Data**

All the data collected or analyzed during the current study are also available from the corresponding author upon reasonable request.

- **Ethical considerations**

Ethical approval was obtained from the Institutional Review Board of the University of Kisangani. All procedures contributing to this work comply with the ethical standards of DRC and the institutional committee on human experimentation and with the Helsinki Declaration of 1975, as revised in 2008.

- **Consent to participate**

We obtained a written informed consent from the participants.

- **Declaration of competing interests**

The authors declare that they have no competing interests.

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