Prevalence of *Plasmodium falciparum* and Haemoglobin Genotype Distribution among Malaria Patients in Zaria, Kaduna State, Nigeria

Gideon Yakusak Benjamin¹*, Benjamin Bartholomew¹, Jabir Abdullahi² and Liman Mubarak Labaran²

¹Department of Agricultural Technology, Nuhu Bamalli Polytechnic Zaria, Nigeria.  
²Department of Science Laboratory Technology, Nuhu Bamalli Polytechnic Zaria, Nigeria.

Authors’ contributions

This work was carried out in collaboration among all authors. Author GYB designed the study, performed the laboratory work, statistical analysis and wrote the first draft of the manuscript. Authors BB, JA and LML managed the analyses and literature searches. All authors read and approved the final manuscript.

Article Information

Received 15 October 2019  
Accepted 18 December 2019  
Published 23 December 2019

ABSTRACT

**Background and Aim:** Malaria remains a disease of public health concern in Nigeria and other parts of Africa. It has caused the death of millions of people; especially pregnant women and children in sub-Saharan Africa. This study was aimed at determining the prevalence of *Plasmodium falciparum* and haemoglobin genotype distribution among malaria patients in Zaria, Kaduna State, Nigeria.

**Methods:** A cross sectional study was carried out in which 300 consenting participants were enrolled; blood samples were collected from them and screened for *Plasmodium falciparum* using Rapid Diagnostic Test (RDT) and microscopy for confirmation. Bio-data and other relevant information were obtained using structured questionnaire and analyzed statistically. The haemoglobin (Hb) genotypes of all the malaria positive patients were determined.

**Results:** A total prevalence of 21.7% was obtained in this study. Malaria prevalence was higher (29.1%) in participants who were not using insecticides at home compared to those who were using...
insecticide at home (17.4%). The $P$ value was significant and the odds ratio showed a significant positive association ($p=0.018$, OR=0.512, CI=0.294-0.894). Participants with HbAA genotype had the highest percentage of malaria 76.9% (50/65), followed by those with HbAS 18.5% (12/65), HbAC 3.1% (2/65) and HbSS 1.5% (1/65) having the least.

**Conclusion:** Apparently, the use of insecticides to kill mosquitoes around the home helps to reduce malaria prevalence. The haemoglobin genotypes HbAS, HbAC and HbSS may be protective against the development of malaria, as their percentages in this study were relatively low compared to HbAA.

**Keywords:** Malaria; plasmodium; microscopy; haemoglobin; mosquitoes.

1. **INTRODUCTION**

Malaria is still a major public health problem in 97 countries and territories in the tropics and subtropics. Approximately 214 million cases of malaria occur annually and 3.2 billion people are at risk of infection globally [1]. Malaria is a parasitic disease caused by *Plasmodium* parasites. The parasites are spread to people through the bites of infected female *Anopheles* mosquitoes. There are 5 parasite species that cause malaria in humans, and 2 of these species; *Plasmodium falciparum* and *Plasmodium vivax* pose the greatest threat [2]. In a non-immune individual, symptoms usually appear 10–15 days after the infective mosquito bite. The first symptoms; fever, headache, and chills; may be mild and difficult to recognize as malaria. If not treated within 24 hours, *P. falciparum* malaria can progress to severe illness, often leading to death [2].

In 2017, nearly half of the world’s population was at risk of malaria. Most malaria cases and deaths occur in sub-Saharan Africa [3]. According to the world malaria report, released in November 2018, there were 219 million cases of malaria in 2017, up from 217 million cases in 2016 [2].

Haemoglobin S (HbS) has become a stable polymorphism within malaria endemic regions, associated with a limited life expectancy among homozygous individuals who suffer from sickle cell disease, and an extended life expectancy of heterozygous individuals who are more likely to evade malaria [4]. HbAS is widely known to confer significant protection from severe and uncomplicated malaria [5] although underlying mechanisms are not precisely defined.

The rate of malaria infection is higher in pregnant women because of their decreased immunity. Many pregnant women living in areas of low or unstable malaria transmission have little or no immunity to malaria and are at higher risk of developing the severe disease as a result of malaria infection than non-pregnant adults living in the same area. Pregnant women with malaria have an increased risk of abortion, stillbirth, premature delivery, and low birth weight infants [6]. Moreover, in unstable malaria transmission areas, pregnant mother’s death may be due to complications of severe malaria (hypoglycaemia, cerebral malaria, and pulmonary edema) or indirectly from malaria-related severe anemia [7].

The most substantive malaria prevention and control measures currently in use include insecticide-treated bed nets (ITNs), indoor residual spraying (IRS), intermittent preventive treatment for pregnant women (IPTp), seasonal malaria chemoprevention (SMC), and diagnosis by malaria microscopy or rapid diagnostic test (RDT), together with effective treatment for confirmed malaria cases with artemisinin-based combination therapies (ACTs) [8]. This study was aimed at determining the prevalence of *Plasmodium falciparum* and haemoglobin genotype distribution among malaria patients in Zaria, Kaduna State, Nigeria.

2. **MATERIALS AND METHODS**

2.1 Study Area

The study was conducted at Hajiya Gambo Sawaba General Hospital, Zaria, Kaduna State, Nigeria. Zaria is a major city in Kaduna state, Nigeria, as well as being a Local Government Area. It is located at 11.11 latitude and 7.72 longitude and it is situated at elevation 644 meters above sea level. Zaria has a population of 975,153 making it the second largest city in Kaduna [9].

2.2 Study Design and Sample Size

The study was a cross sectional study that lasted for 6 months (from January to June 2019). All febrile patients presenting symptoms of malaria,
who were directed to the laboratory for malaria parasite (MP) test, were included, while patients directed to the laboratory for laboratory tests other than MP test were excluded. A sample size of 267 was obtained using a previous prevalence of 22.4% [10] in Kaduna State, and the formula described by Naing et al. [11]. However, a total of three hundred (300) blood samples were collected for the study.

2.3 Administration of Structured Questionnaire and Sample Collection

A structured questionnaire was administered to individuals who met the inclusion criteria after obtaining their consent to participate in the study. This was used to obtain bio-data and other information relevant to the research. Venipuncture technique was used to collect 2ml of blood which was transferred into an EDTA container and later screened for Plasmodium falciparum.

2.4 Screening of Blood Samples for Plasmodium falciparum

Blood samples were screened by CareStart™ Malaria HRP2 Rapid Diagnostic Test (RDT) Kit (Access Bio, Inc, Somerset, NJ), specific for the detection of Plasmodium falciparum. The RDT test was carried out according to manufacturer’s instructions and positive samples were confirmed by microscopic examination of Giemsa stained thick and thin blood smears using the technique described by Cheesbrough [12].

2.5 Determination of Haemoglobin Genotype

Cellulose acetate method of haemoglobin electrophoresis was carried out as described by Egesie et al. [13] to determine the haemoglobin genotype of the Plasmodium positive samples.

3. RESULTS

Table 1 shows the prevalence of malaria in Hajiya Gambo Sawaba Hospital, Zaria. Out of the 300 samples examined, 65 were positive with Plasmodium falciparum while the remaining 235 were negative. A total prevalence of 21.7% was obtained.

The prevalence of malaria in relation to pregnancy is shown in Table 2. Out of 300 study participants, 185 were females from which 35 were pregnant while the remaining 150 were not.

There was lower prevalence of Plasmodium falciparum infection (14.3%) in those who were pregnant compared to those who were not pregnant (22.7%). The p value and odds ratio were not significant (P=0.274, OR=0.569, 95% CI=0.205-1.578).

Table 1. Plasmodium falciparum prevalence in Hajiya Gambo Sawaba Hospital, Zaria

<table>
<thead>
<tr>
<th>Malaria status</th>
<th>Number examined</th>
<th>% Prevalence</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>65</td>
<td>21.7</td>
</tr>
<tr>
<td>Negative</td>
<td>235</td>
<td>78.3</td>
</tr>
<tr>
<td>Total</td>
<td>300</td>
<td>100</td>
</tr>
</tbody>
</table>

Table 3 shows the prevalence of malaria in relation to ITN use at home. Participants who were using ITN at home had a higher prevalence (27.0%) than those who were not (19.9%). The Odds ratio and P value showed no significant association (P=0.197, OR=0.671, CI=0.365-1.233).

The prevalence of malaria in relation to insecticide use at home is shown in Table 5. We found higher malaria prevalence (29.1%) in participants who were not using IRS at home than those who were (17.4%). The P value was significant and the odds ratio showed a significant positive association (P=0.018, OR=0.512, CI=0.294-0.894).

Figure 1 shows the distribution of malaria based on haemoglobin genotype of malaria positive participants. Participants with HbAA genotype had the highest percentage of malaria 76.9% (50/65), followed by those with HbAS 18.5% (12/65), HbAC 3.1% (2/65) and HbSS 1.5% (1/65).

4. DISCUSSION

Three hundred (300) participants were enrolled in this study and screened for Plasmodium falciparum. The parasite was detected in 65 of the participants, and a total prevalence of 21.7% was obtained in this study. This is lower than previous malaria prevalence of 22.4% and 25.3% reported by Aliyu et al. [10] and Okogwu et al. [14] respectively in Kaduna State. The reduction in malaria prevalence may be as a result of the use of malaria preventive measures such as insecticide spray at home.

In this study, participants who were spraying their homes with insecticide to kill mosquitoes had lower malaria prevalence than those who were not doing so. The World Health Organization [15]...
reported that the number of people protected as a result of indoor residual spray with IRS usage increased from 13 million in 2005 to 75 million in 2009. Some field studies have reported the effectiveness of IRS in reducing malaria prevalence [16]. Aspects of geography, entomology, human behavior, and community acceptance of the IRS program could contribute to why IRS is more successful in one community than in another. Several studies have also reported a similar finding [17,18].

The heterozygous state for hemoglobin S [HbAS] is the best described of all malaria-protective traits and is used as the classic example of balanced polymorphism in schools and colleges throughout the world [19]. The close link between the geographic distribution of HbAS and that of malaria, documented by a number of different investigators years ago [20,21,22] gave the first clues. However, a more definite hypothesis of malaria with respect to HbAS was articulated by Allison [23,24]. This relationship has been quantified statistically, an analysis that found strong geographical support for the malaria hypothesis in Africa but not in either the Americas or in Asia [25]. After determining the haemoglobin genotypes of all the Plasmodium falciparum positive participants in the current study, we realized that majority of the participants had the haemoglobin genotype AA (HbAA). The prevalence of participants with HbAS and HbAC were relatively low compared to HbAA. This is similar to the reports of Albiti and Nsiah [26] and Akanbi [27]. According to Akanbi [27], haemoglobin genotype influences the prevalence of malaria in endemic areas. The level of susceptibility to malaria infection has been reported to be higher in individuals with HbAA when compared to those with HbAS and HbAC, thus, the high frequency of HbAC and HbAS in malaria endemic areas has been attributed to a decrease in malaria morbidity and mortality in malaria endemic areas [28]. A similar finding was reported by Aidoo [29]. Williams and Weatherall [19] reported that HbAS is associated with strong protection against all forms of clinical falciparum malaria. In their study, Williams et al. [30] also reported that HbAS was 90% protective against the development of severe or complicated malaria. Earlier studies attributed this protection to impairment in the invasion and growth of P. falciparum parasites into HbAS red cells under conditions of low oxygen tension that were physiologically representative of in vivo conditions [31,32,33]. In this study participants with HbSS (sicklers) had the least prevalence of malaria. This is in agreement with the reports of Allison [23, 24, 34] who noted that “sicklers” were significantly less likely than “non-sicklers” to carry malaria parasites under conditions of natural exposure. He also showed that, when inoculated intravenously with large volumes of parasite-infected blood, sicklers were less likely to develop clinical malaria. Whereas the malaria-protective effect of HbAS is clear, the effect of the homozygous state, HbSS, on malaria risk is controversial [19].

![Haemoglobin genotype](image)

Fig. 1. Percentage of malaria based on haemoglobin genotype of malaria positive participants

Table 2. Prevalence of malaria in relation to pregnancy

<table>
<thead>
<tr>
<th>Pregnant</th>
<th>No. examined</th>
<th>No. positive</th>
<th>Prevalence</th>
<th>$\chi^2$</th>
<th>P value</th>
<th>OR</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>35</td>
<td>5</td>
<td>14.3</td>
<td>1.198</td>
<td>0.274</td>
<td>0.569</td>
<td>0.205-1.578</td>
</tr>
<tr>
<td>No</td>
<td>150</td>
<td>34</td>
<td>22.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>185</td>
<td>39</td>
<td>21.1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Key: OR=Odds ratio, CI=Confidence interval
Table 3. Prevalence of malaria in relation to ITN use at home

<table>
<thead>
<tr>
<th>ITN use</th>
<th>No. examined</th>
<th>No. positive</th>
<th>Prevalence</th>
<th>$\chi^2$</th>
<th>P value</th>
<th>OR</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>226</td>
<td>45</td>
<td>19.9</td>
<td>1.663</td>
<td>0.197</td>
<td>0.671</td>
<td>0.365-1.233</td>
</tr>
<tr>
<td>No</td>
<td>74</td>
<td>20</td>
<td>27.0</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>300</td>
<td>65</td>
<td>21.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Key: ITN= Insecticide Treated Net, OR=Odds ratio, CI=Confidence interval

Table 4. Prevalence of malaria in relation to the use of insecticide at home

<table>
<thead>
<tr>
<th>INS use</th>
<th>No. examined</th>
<th>No. positive</th>
<th>Prevalence</th>
<th>$\chi^2$</th>
<th>P value</th>
<th>OR</th>
<th>95%CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Yes</td>
<td>190</td>
<td>33</td>
<td>17.4</td>
<td>5.641</td>
<td>0.018</td>
<td>0.512</td>
<td>0.294-0.894</td>
</tr>
<tr>
<td>No</td>
<td>110</td>
<td>32</td>
<td>29.1</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>300</td>
<td>65</td>
<td>21.7</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Key: INS=Insecticide, OR=Odds ratio, CI=Confidence interval

Few years back, Gouagna and colleagues working in Burkina Faso, made an additional intriguing observation regarding the malaria-protective effect of HbS [35]. Although HbAS protects against clinical malaria infections, parasite-infected blood from HbAS subjects is several times more infectious to the Anopheles vector than that of normal subjects [35]. This observation, which is echoed by a similar observation from Senegal [36], suggests that the personal advantage of HbAS is not only balanced by the cost of potential homozygosity in offspring but also by that of increased malaria transmission to the general population [19]. In this study, only 3.1% of those with malaria had HbAC genotype, this is relatively low compared to the percentage of HbAA. Taylor et al. [37] reported that homozygotes for HbC i.e. HbCC, were strongly protected against severe malaria, and heterozygotes (HbCC) were mildly protected. This could be why he had such a small percentage in this study.

5. CONCLUSION

The use of IRS at home was found to be significantly associated with reduced *Plasmodium falciparum* infection, and the percentage of participants with malaria was higher among those with HbAA genotype compared to other genotypes examined in this study.

DISCLAIMER

This paper is based on preliminary dataset. Readers are requested to consider this paper as preliminary research article, as authors wanted to publish the initial data as early as possible. Authors are aware that bigger sample size is required to get a scientifically established conclusion. Readers are requested to use the conclusion of this paper judiciously as authors have worked with a small sample size. Authors also recommend working with bigger sample size for similar future studies.

CONSENT

As per international standard, participants' written consent has been collected and preserved by the author(s).

ETHICAL APPROVAL

Full ethical approval was obtained from the Health Research Ethical Committee (HREC) of Kaduna State Ministry of Health and Human Services.

ACKNOWLEDGEMENT

We appreciate TETFUND Nigeria for providing financial support for this research.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

17. Hamusse SD, Balcha TT, Belachew T. The impact of indoor residual spraying on malaria incidence in East Shoa Zone, Ethiopia. Global Health Action. 2012; 5. DOI: 10.3402 / gha.v5i0.11619


