

## Research Article

# Asymptomatic Malaria Correlates with Anaemia in Pregnant Women at Ouagadougou, Burkina Faso

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Sub-Saharan Africa records each year about thirty-two million pregnant women living in areas of high transmission of *Plasmodium falciparum* causing malaria. The aim of this study was to carve out the prevalence of asymptomatic malaria among pregnant women and to emphasize its influence on haematological markers. The prevalence of *Plasmodium falciparum* asymptomatic infection among pregnant women was 30% and 24% with rapid detection test (RDT) and microscopy, respectively. The prevalence of *P. falciparum* asymptomatic malaria was reduced among pregnant women using sulfadoxine-pyrimethamine's intermittent preventive treatment and 61% of them were anaemic. Anaemia was significantly more common in women infected with *P. falciparum* compared with the uninfected pregnant women. Most of the women had normal levels of homocysteine and low levels of folate, respectively. Therefore, the systematic diagnosis of malaria should be introduced to pregnant women as a part of the antenatal care.

## 1. Introduction

Sub-Saharan Africa records each year about thirty-two million pregnant women living in areas of high transmission of *Plasmodium falciparum* causing malaria [1]. In malaria-endemic areas, malaria is the cause for almost 25% of maternal deaths each year [2], with the greatest risk of infection and morbidity occurring in primiparous women, adolescents, and those coinfecting with HIV.

Malaria is still the leading cause of medical consultation in Burkina Faso. In 2009, malaria accounted for 45% of medical consultation, 57% of hospitalization, and 51% of mortality. The number of clinical cases of malaria recorded in health care facilities was 4,507,420 including 11,810 deaths. Malaria's morbidity affected 15,394 pregnant women with 172 deaths among them [3].

A previous study demonstrated that asymptomatic malaria in pregnant women induces maternal anaemia and

low birth weight infants [4]. In areas of malaria transmission such as Burkina Faso, infection with *P. falciparum* in pregnant women may be asymptomatic but accompanied by secondary anaemia and a placental parasitaemia.

In Burkina Faso, many studies have investigated the course of acute malaria during pregnancy [5–7], but there are only few data on asymptomatic malaria in pregnant women.

The purpose of this study was to evaluate the prevalence of asymptomatic malaria among pregnant women. Our study also assessed the relationship between pregnant women's biochemical and haematological parameters and asymptomatic malaria.

## 2. Materials and Methods

**2.1. Population.** The study was conducted at Saint Camille Medical Centre (CMSC) in Ouagadougou during the malaria transmission season from September until October 2010.

Two hundred and one (201) primigravida and multigravida without any prior evidence of clinical malaria were recruited into the study. After obtaining consent, up to 5 mL of their venous blood was collected. Asymptomatic malaria infection was assessed by rapid detection test Acon (ACON Malaria Pf, San Diego, USA) and microscopic examination of Giemsa-stained thick and thin blood smears. All slides were read twice by experienced microscopists and discrepancies resolved by a third reader (limit of detection, approximately 2 parasites/mL) [8]. The study was approved by the Joint CERBA/Saint Camille Ethics Committee.

**2.2. Haematological and Biochemical Parameters of Pregnant Women.** ABO and Rh blood groups determinations were carried out on a slide using monoclonal blood grouping antisera; anti-A, anti-B, anti-AB, and anti-D (BIOTEC Laboratories Ltd., Great Britain).

Haemoglobin and serum iron levels were performed using two colorimetric kits (Cypress DIAGNOSTICS, Langdorp, Belgium) following the manufacturer's instructions. Haemoglobin typing was performed by cellulose acetate electrophoresis.

Dosages of folate and homocysteine were made using AxSYM kits for folate and homocysteine (Abbott, Santa Clara, CA, USA) following the manufacturer's instructions.

**2.3. Statistical Analysis.** Data were entered and analyzed using SPSS version 17 statistical package and EPI-Info version 6.04 dfr (CDC, Atlanta, USA). Differences in the prevalence of asymptomatic malaria for sociodemographic, haematological, and biochemistry variables were tested for significance using chi square tests. *P* value less than 0.05 was considered statistically significant.

### 3. Results

**3.1. Sociodemographic Characteristics of Pregnant Women.** Two hundred and one women (201) agreed to participate in the study. Pregnant women were aged from 18 to 39 years with a mean age of  $25 \pm 5$  years. Most of the pregnant women belonged to an age group under 30 years (78%). Women were from different professional groups; the majority was housewives (61%), illiterate (42%), and multiparous (65%). Only 42% of all pregnant women were using an insecticide-treated net (ITN) and 40% were taking an iron supplement as shown in Table 1.

**3.2. Prevalence of *Plasmodium falciparum* Asymptomatic Malaria in Pregnant Women.** The prevalence of *Plasmodium falciparum* asymptomatic infection among pregnant women was 30% and 24% with rapid detection test (RDT) and microscopy, respectively.

The Acon rapid test had a sensitivity of 100% and a specificity of 92% for the detection of *Plasmodium falciparum* asymptomatic malaria compared to microscopic diagnosis. The mean parasite density observed in pregnant women was 4058 parasites/ $\mu$ L.

TABLE 1: Sociodemographic characteristics of pregnant women.

| Characteristics            | Number | Percentage (%) |
|----------------------------|--------|----------------|
| Profession                 |        |                |
| Housewife                  | 123    | 61             |
| Informal sector            | 60     | 30             |
| Employee                   | 5      | 2              |
| Student                    | 13     | 7              |
| Education level            |        |                |
| Illiterate                 | 84     | 42             |
| Primary                    | 59     | 29             |
| Postprimary                | 58     | 29             |
| Age (years)                |        |                |
| $\leq 20$                  | 50     | 25             |
| 21–25                      | 59     | 29             |
| 26–30                      | 59     | 29             |
| 31–39                      | 33     | 16             |
| Number of children         |        |                |
| 1                          | 70     | 35             |
| 2                          | 45     | 22             |
| $\geq 3$                   | 86     | 43             |
| Stage of pregnancy         |        |                |
| 1st trimestre              | 70     | 35             |
| 2nd trimestre              | 96     | 48             |
| 3rd trimestre              | 35     | 17             |
| Use of impregnated bednet  |        |                |
| Yes                        | 85     | 42             |
| No                         | 116    | 58             |
| Use of iron and folic acid |        |                |
| Yes                        | 81     | 40             |
| No                         | 120    | 60             |

**3.3. Haematological and Biochemical Parameters in Pregnant Women.** As shown in Table 2, the average of haemoglobin level in pregnant women was  $10.5 \pm 1.7$  g/dL. The majority of pregnant women (61%) had a haemoglobin level of  $<11$  g/dL and a normal level of serum iron (83%). Homocysteine and folate levels were also measured in pregnant women. Most of the women (90% and 62%) had normal levels of homocysteine ( $\leq 12$  mmol/L) and low levels of folate ( $<7.7$  ng/mL), respectively.

We found that anaemia was significantly more common in women infected with *P. falciparum* (33%) compared with the uninfected pregnant women (10%) ( $P = 0.0002$ ) (Table 3).

We did not observe statistically significant differences between both gravida and asymptomatic malaria and the use of insecticide-treated nets and *P. falciparum* asymptomatic malaria. However, the presence of *P. falciparum* was significantly reduced in pregnant women who had received intermittent preventive treatment based on sulfadoxinepyrimethamine (SP) (13%) compared to those without treatment (29%) ( $P = 0.0182$ ) (Table 3). Among the pregnant women, the prevalence of HbAS (8%), HbAC (16%), HbCC (1%),

TABLE 2: Hematological and biochemistry parameters of pregnant women.

| Parameters                               |                           | Normal values | Number | Percentage |
|--|---------------------------|---------------|--------|------------|
| Haemoglobin level<br>(10.49 ± 1.73 g/dL) | Severe anaemia            | <7            | 2      | 1          |
|  | Moderate anaemia          | [7–10[        | 74     | 37         |
|  | Slight anaemia            | [10–11[       | 47     | 23         |
|  | Absence of anaemia        | ≥11           | 78     | 39         |
| Serum iron<br>(14.64 ± 1.96 μmol/L)      | Low                       | <7.16         | 21     | 10         |
|  | Normal                    | 7.16–26.85    | 167    | 83         |
|  | High                      | >26.85        | 13     | 7          |
| Folate<br>(7.01 ± 2.84 ng/mL)            | Low                       | <7.7          | 125    | 62         |
|  | Normal                    | ≥7.7          | 76     | 38         |
| Homocysteine<br>(10.49 ± 1.73 μmol/L)    | Normal                    | ≤12           | 181    | 90         |
|  | Moderate Homocysteinaemia | ]12–30[       | 18     | 9          |
|  | Hyperhomocysteinaemia     | ≥30           | 2      | 1          |

TABLE 3: Prevalence of *P. falciparum* in pregnant women according to their hematological and sociodemographic status.

| Parameters                   |  | Blood smears  |              | P value       |
|------------------------------|--|---------------|--------------|---------------|
|                              |  | Negative      | Positive     |               |
| Haemoglobin level            | Presence of anaemia<br>N = 123/201 (61%) | 82/123 (67%)  | 41/123 (33%) | <b>0.0002</b> |
|                              | Absence of anaemia<br>N = 78/201(39%)    | 70/78 (90%)   | 8/78 (10%)   |               |
| Stage of pregnancy           | 1st trimestre<br>N = 70/201 (35%)        | 55/70 (79%)   | 15/70 (23%)  | NS            |
|                              | 2nd trimestre<br>N = 96/201 (48%)        | 68/96 (71%)   | 28/96 (29%)  |               |
|                              | 3rd trimestre<br>N = 35/201 (17%)        | 29/35 (83%)   | 6/35 (17%)   |               |
| Number of pregnancy          | 1<br>N = 70/201 (35%)                    | 53/70 (76%)   | 17/70 (24%)  | NS            |
|                              | 2<br>N = 45/201 (22%)                    | 34/45 (76%)   | 11/45 (24%)  |               |
|                              | ≥3<br>N = 86/201 (43%)                   | 65/86 (76%)   | 21/86 (24%)  |               |
| Preventive treatment<br>(SP) | Yes<br>N =146/201 (73%)                  | 104/146 (71%) | 42/146 (29%) | <b>0.0182</b> |
|                              | No<br>N = 55/201 (27%)                   | 48/55 (87%)   | 7 /55 (13%)  |               |
| Haemoglobin type             | AA (74%)                                 | 74%           | 26%          | NS            |
|                              | AC (16%)                                 | 81%           | 19%          |               |
|                              | AS (8%)                                  | 75%           | 25%          |               |
|                              | CC (1%)                                  | 1/1           | 0/1          |               |
|                              | SC (1%)                                  | 2/2           | 0/2          |               |
| Blood groups                 | A (25%)                                  | 72%           | 28%          | NS            |
|                              | B (31%)                                  | 73%           | 27%          |               |
|                              | AB (9%)                                  | 71%           | 29%          |               |
|                              | O (35%)                                  | 82%           | 18%          |               |

NS: not significant; in bold: significant P values.

and HbSC (1%) hemoglobin types was not associated with asymptomatic malaria (Table 3).

We found that pregnant women from blood group O were less infected by *P. falciparum* than those belonging to the non-O blood groups but the difference was not statistically significant.

#### 4. Discussion

This work aimed to study asymptomatic malaria among pregnant women consulting Saint Camille Medical Centre at Ouagadougou in Burkina Faso. We have shown that the prevalence of asymptomatic malaria among women was 30% and 24% using an RDT and microscopy, respectively. The difference was not significant but could be explained by the persistence of histidine rich protein 2 (HRP-2) antigen of *P. falciparum* after the parasite clearance [9]. The RDT used had a sensitivity of 100% and a specificity of 92% compared to conventional microscopy. Our results are comparable to those from studies in Cote d'Ivoire and Senegal with sensitivities of 100% and 96% and specificities of 88% and 87%, respectively, [10, 11].

We evaluated through a questionnaire the use of ITNs in pregnant women. Only 42% of the women used an ITN. Several reasons explain the low use of ITNs: psychological effects of suffocation, ignorance of the risks of malaria during pregnancy, and the cost of the net. Utilization of ITNs varies between studies, 58% and 9% in Burkina [6, 12] and 16% in Nigeria [13].

The fight against malaria in pregnant women involves the use of insecticide-treated nets and intermittent preventive treatment (IPT) with sulfadoxine-pyrimethamine (SP). Thus, all pregnant women living in areas of stable malaria transmission should receive at least two doses of IPT as recommended by the World Health Organization [14].

Only 27% of the pregnant women have taken the first dose of SP and less than half of the women used iron supplement and folic acid. This low use of iron and SP can be explained by the fact that most of the women began their prenatal checks from the fifth month of the pregnancy for various reasons including the ignorance of risk associated with poorly managed and monitored pregnancy and a lack of financial resources.

The use of SP as an intermittent preventive treatment by pregnant women was associated with a reduced prevalence of *P. falciparum* asymptomatic infection compared to those who did not receive SP (13% versus 29%,  $P = 0.0182$ ). Our results are comparable to those obtained in Ghana [15] where 15% versus 45% of *P. falciparum* infection was observed among pregnant women in the presence or absence of the intermittent preventive treatment, respectively.

WHO defined anaemia in pregnant women as a haemoglobin level  $<11$  g/dL [16]. We found that 61% (123/201) of the pregnant women in the study were anaemic. Interestingly, this anaemia was correlated with the presence of *P. falciparum* asymptomatic. Indeed, 33% of all pregnant anaemic women were positive for *P. falciparum* compared with 10% of anaemic women without parasitaemia. The prevalence of anaemia observed in this study is comparable

to those of previous studies obtained 66% [17], 64% [12] in Burkina Faso, and 64% [18] in Benin. The correlation between *P. falciparum* and anaemia found in this study is similar to those reported in Nigeria [19, 20].

We did not find a significant correlation between anaemia and gravida. This is in accordance with a previous report on *P. falciparum* asymptomatic malaria in Nigerian pregnant women [21]. However, other studies have shown higher rates of anaemic women in primiparous women compared to multiparous ones [19, 22, 23].

Nutritional deficiencies including iron and folic acid have been linked to anaemia. Our study shows that 83% of pregnant women had a normal serum iron but 62% of them had a low level of folate; this is in discordance with what has been previously reported on Nigerian pregnant women [24].

High levels of homocysteine (HCY) play a role in pregnancy complications such as an increased risk of eclampsia, spina bifida malformation, children with low birth weight, stillbirths, and premature births [25]. Most pregnant women (90%) had normal values of homocysteine ( $\leq 12$   $\mu\text{mol/L}$ ). The mean value of homocysteine (10.5  $\mu\text{mol/L}$ ) found in pregnant women is higher than the value of 3.6  $\mu\text{mol/L}$  obtained in healthy women in Burkina Faso [26]. This difference could be explained by physiological changes during pregnancy. However, the average homocysteine levels found in our study were lower than those found in Nigeria (14.1  $\mu\text{mol/L}$ ) [24] and the difference could be due to the high prevalence of moderate hyperhomocysteinaemia among adolescents and adults of both sexes in the Nigerian population [27]. Indeed studies conducted in Burkina showed that plasma homocysteine (HCY) varies according to gender, age, and nutritional habits [26, 28]. We did not find an association between haemoglobin type and anaemia in pregnant women. With regard to blood groups, we found that pregnant women of blood group O were less infected than those of non-O blood groups, but this difference was not significant. However, that study in Nigeria found that pregnant women group O were more infected by *P. falciparum* [4].

Our findings are interesting because we have demonstrated as previously reported in other studies [4, 29] that *P. falciparum* asymptomatic malaria is responsible for anaemia in pregnant women in Burkina Faso. However, there are some potential limitations in our study to be considered. First, *P. falciparum* asymptomatic malaria was identified using RDT and optical microscopy. The sensitivity of these tests is low for the detection of very low parasitaemia. Indeed, a recent study has shown a higher prevalence of asymptomatic malaria detected by polymerase chain reaction (PCR) compared with other diagnostic methods [9]. Thus, the prevalence of asymptomatic malaria reported in our study may be underestimated. In addition, to define anaemia we only measured haemoglobin levels among pregnant women.

Several studies documented an impact of asymptomatic malaria on haematological markers like blood count, ferritin, and transferrin levels [30–32]. But our study did not include the evaluation of those parameters.

A subsequent study could focus on the role of folic acid in reducing anaemia among pregnant women with mild or

asymptomatic malaria. Intermittent preventive treatment of malaria in pregnant women is ensured by SP. Therefore, monitoring of SP resistance by *P. falciparum* is necessary among pregnant women.

## 5. Conclusion

This study revealed a high prevalence of asymptomatic malaria among pregnant women in Burkina Faso. This asymptomatic infection is responsible for anaemia in pregnant women and may count in mother to children malaria transmission. Therefore, the systematic diagnosis of malaria should be introduced to pregnant women as a part of the antenatal care. In addition, intermittent preventive treatment of malaria and the use of ITNs should be strengthened among pregnant women in Burkina Faso.

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