



Detection of malaria parasites by microscopy and rapid diagnostic test kit (PLDH) in pregnant women and children, Lagos, Nigeria

Audu, H. O.^{1*} and Abdulsalam, M. Y.²

¹Department of Biological Sciences

Federal University Lokoja, P.M.B 1152 Lokoja, Kogi State Nigeria

²Department of Biological Sciences, Bayero University, Kano, Kano State

email: *halimaa.28@gmail.com*

Abstract

The effectiveness of Rapid Diagnostic Test Kit (RDT) was compared with microscopy for the evaluation of malaria infection in children and pregnant women attending two selected health facilities in Lagos State, south-western, Nigeria. A total of 482 patients comprising 252 pregnant women (mean age: 26.86 ± 4.46 years) and 230 children (mean age 24.31 ± 18.19 months) were sampled between November 2010 and August 2011. The prevalence of infection observed in this study was 30% and 6.7% by microscopy in children and pregnant women respectively while RDT showed prevalence of 24.3% and 4.8% in children and pregnant women respectively. The evaluation of RDT showed high specificity of 95.7% and 98.7% for children and pregnant women respectively and low sensitivity of 71.0% and 52.9% for children and pregnant women respectively. The difference in the prevalence of malaria infection detected by microscopy and RDT was statistically significant ($p=0.000$). This study showed that RDT was not as sensitive as microscopy in the detection of malaria infection in the study-area.

Keywords: malaria, microscopy, RDT, pregnant women, Nigeria.

Introduction

Malaria is the leading cause of mortality and morbidity in tropical Africa with children and pregnant women bearing the brunt of the infection [1]. The disease is also responsible for 20% low birth weight and 100,000 infant deaths per year in Africa [2]. The protection of pregnant women living in endemic countries has been of particular interest to many national malaria control programmes because of their low immunity. In endemic areas, acquired immunity though established is liable to break down under the condition of stress in pregnancy [3].

Nigeria has accounted for 25% and 30% of infant and child deaths respectively and 11% maternal mortality. [4] World Health Organisation recommended prompt parasite based diagnosis in

all patients suspected of malaria before treatment is administered. [5] The major drawback in routine microscopy in malaria studies is the expertise in the parasite identification. As a result of this, the use of Rapid Diagnostic Test (RDT) to ensure prompt and early diagnosis as a step to the control of malaria was advocated as a necessity for active surveillance [1 and 6].

However, prompt parasitological confirmation of malaria before treatment is unrealistic in many settings because of dearth of efficient laboratory diagnostic services and/or unavailability of qualified laboratory scientists. The provision of cheap and accurate rapid point of care tests that require little skill is therefore appealing [7]. This study therefore aimed to compare the efficiency of RDT and

microscopy for the detection of malaria infection in children and pregnant women in Lagos, south-western Nigeria.

Materials and method

Study-area and population

The study was conducted in Lagos metropolis, south-western Nigeria. Lagos is located on Latitude 6.4512°N and Longitude 3.3973° E. A total of 482 consented participants (230 young children of 0-5 years and 252 pregnant women of 19-38 years) attending Egbeda Medical Centre and Motayo Hospital, Ikeja, Lagos State, Nigeria from November 2010 to August 2011, were recruited for the study. Structured questionnaires were personally administered to the participants or the care-givers/guardians during the study to obtain information on the individual's history of malaria infection. Ethical clearance was obtained from the authorities of health facilities used for the study.

Blood sample collection

The blood specimen was collected by venous puncture in pregnant women and put into the collection tube (containing EDTA to prevent coagulation). In children, blood sample was taken by finger pricking. Both thin and thick films were prepared from the blood samples collected while the rest samples were used for Rapid Diagnostic Test (RDT).

The films were allowed to dry and stained immediately. Thick film was stained with 3% Giemsa for 40 minutes and thin film with 10% Giemsa. The films were examined using the oil immersion at x100 objective lens.

Rapid Diagnostic Test (RDT)

The *Plasmodium* lactate dehydrogenase assay was performed by dropping a drop of assay diluent in the lysis well and 20 μ l of blood specimen was added and mixed with the capillary pipette. This lysed the red blood cells and released the PLDH. After a minute, 10 μ l of lysed blood was taken from the lysis well and put in the square shaped sample well. Four drops of the assay diluent was added in the other round shaped well. The specimen migrated to the top of the strip as the assay diluents ran through the test strip. After 15 minutes, bands appeared to show evidence of reaction between the sample and the test kit.

The results from the PLDH assay were compared with those of microscopic examination to confirm the sensitivity and specificity of the kit.

Data analysis

The data was analysed using Epi-info 2002 software programme. For paired and unpaired comparisons, ANOVA and *chi*-square analyses were used. 95% confidence interval was applied for the analysis so that p -value less than 0.005 were considered significant. Epi-info 2002 was also used to determine the Specificity, Sensitivity, Positive Predictive Value (PPV) and Negative Predictive Value (NPV).

Results

A prevalence of 30% and 6.7% was recorded by microscopy in children and pregnant women respectively while RDT showed prevalence of 24.3% and 4.8% in children and pregnant women respectively (Table 1).

In children, the sensitivity of RDT was evaluated to be 71.0% and specificity as 95.7%. The Negative Predictive Value (NPV) was 88.5% and Positive Predictive Value (PPV) 87.5% (Table 2).

Pregnant women showed sensitivity of 52.9% and specificity as 98.7%. Negative Predictive Value was 96.7% and the Positive Predictive Value was 75.0%. There was a significant difference between microscopy and RDT ($p=0.000$) (Table 3). The lowest detection limit among children was 440 per micro liter. There was detection by RDT at zero level of parasitaemia for 7 cases which showed false positive and 20 cases of false negative. In pregnant women, lowest detection limit was 2,800 per micro litre. There was detection by RDT at zero level of parasitaemia for 3 cases indicative of 3 cases of false positive and 8 cases of false negative.

Table 1. Prevalence of malaria in children and pregnant women.

	Children		Pregnant Women	
	No of Negative	No of Positive	No of Negative	No of Positive
Microscopy	161 (70%)	69 (30.0%)	235 (93.3%)	17 (6.7%)
RDT	174 (75.7%)	56 (24.3%)	240 (95.2%)	12 (4.8%)

Key: RDT: Rapid Diagnostic Test.

Table 2. Sensitivity and specificity in children.

Malaria	RDT Grouping		Total
	Negative	Positive	
Negative	154 (95.7%)	7 (4.3%)	161 (70%)
Positive	20 (29.0%)	49 (71.0%)	69 (30.0%)
Total	174 (75.7%)	56 (24.3%)	230 (100%)

Sensitivity = 71.0%.

Specificity = 95.7%.

Negative Predictive Value (NPV) = 88.5%.

Positive Predictive Value (PPV) = 87.5%.

Table 3. Sensitivity and specificity in pregnant women.

Malaria	RDT Grouping		Total
	Negative	Positive	
Negative	232 (98.7%)	3 (1.3%)	235 (93.3%)
Positive	8 (47.1%)	9 (52.9%)	17 (6.7%)
Total	240 (95.2%)	12 (4.8%)	252 (100%)

Sensitivity = 52.9%.

Specificity = 98.7%.

Negative Predictive Value (NPV) = 96.7%.

Positive Predictive Value (PPV) = 75.0%.

Discussion

Malaria is a major cause of death in young children and pregnant women [5]. This is due to severe complications such as anaemia and untimely death caused especially in the case of *P. falciparum* [8]. Early detection of an infection is crucial to prompt treatment. Diagnosis of malaria using microscopy could be delayed by lack of manpower, electricity equipment and many other reasons [9]. The results of RDT revealed the prevalence rate of 24.3% and 4.8% in children and pregnant women respectively while microscopy revealed 30% and 6.7% in children and pregnant women respectively. The prevalence observed in the present study using both methods was extremely low when compared with 82% prevalence reported in pregnant women in Osogbo [8], but compared favourably with the recent reports of [1 and 9] in Lagos State where a prevalence less than 40% was recorded in both studies. This temporal and spatial variation in malaria in different parts of Nigeria could be premised on many factors. These among many others include the environmental and climatic factors prompting mosquito vectors, vectors competency and availability of preventive measures. [9] [1] also speculated the problem of misdiagnosis in places where high malaria prevalence has been recorded.

Comparison between microscopy and RDT kit showed 71.0% sensitivity in children and 52.9% in pregnant women while specificity was 95.5% in children and 98.7% in pregnant women. These observations were in consonance with the report of [10] but at variance with previous reports of [11] and [12] who recorded 100% specificity.

[4] Showed that the RDT is more effective for the diagnosis of malaria in Nigeria. The prevalence rate reported in pregnant women and children by both methods in this study were less than 40% as it was the case for other reports [1 and 9]. Moreover, the sensitivity obtained by using RDT did not meet the criteria for detecting at least 100 parasite per micro litre of blood as stated by [13 and 12] this low sensitivity obtained remains a disadvantage because parasitaemia lower than 100 parasites can still be detected by microscopy. False negative result in the study could be attributed to low parasitaemia which may not be detected by RDT [12] the consequence of false positive is the use of unnecessary treatment for patients not suffering from malaria which could later lead to drug pressure and resistance. Also the false negative result could eliminate a malaria infected patient from treatment and later lead to death [14].

Conclusion

The results showed that the RDT is less efficient for the diagnosis of malaria in pregnant women and children in Lagos. This plausibly suggests that the RDT may be of less significant value in the diagnosis of malaria infection in Lagos State. This in turn suggests that microscopy still remain the gold standard for the malaria diagnosis. Therefore, there was need for constant training of laboratory technicians for effective diagnosis of malaria using microscopy.

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