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Comparative Use of RDT and Thick Film Microscopy in the Diagnosis of Malaria in Sub-urban Settlements in Makurdi, Nigeria

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Abstract: In as much as accurate diagnosis seems to be the only way of effecting rational therapy, it has been the most neglected area of malaria research. Thick blood film microscopy and Rapid diagnostic test (RDT) was comparatively used to study malaria prevalence in Makurdi, Nigeria. A total of 328 blood samples were collected from consented respondents and analyzed using Blood film examination with Field's stains A and B staining techniques and Care Start [TM] rapid diagnostic test (RDT) manufactured by Access Bio Inc, USA to detect the presence of malaria parasites in blood. Questionnaires were used to get demographics of the respondents. Of the 328 participants examined, 164(50.0%) were positive for malaria parasites by light microscopy and 32(9.8%) were positive for malaria by RDT Care Start [TM] HRP2. The sensitivity and specificity of RDT was found to be 16.5% and 97.0% respectively while the Positive Predictive Value (PPV) and Negative Predictive Value (NPV) was found to be 84.4% and 53.7% respectively. The females 92(51.7%) and 20(11.2%) were slightly more infected than the males 72(48.0%) and 12(8.0%) using both methods, but result was not statistically significant (P>0.05). Malaria prevalence was higher among those with no formal education and least among those with tertiary education (P>0.05). The prevalence of malaria in respect to location was significantly higher in New GRA by RDT and Chile by light microscopy and least in High level and Agwan Jukum in order of RDT and microscopy (P=0.000). This study revealed that malaria can affect all sexes irrespective of their educational cadre and location. The Care Start [TM] RDT showed very poor sensitivity in contrast to light microscopy. In as much as light microscopy in poor set ups cannot be used routinely, the RDT has not proven to be a good replacement.

Keywords: Malaria, Makurdi, RDT, Microscopy

1. Introduction

Malaria is one of the highest killer diseases affecting most tropical countries especially Africa. It affects over 500 million people worldwide, killing over one million children annually [1]. Of all the human malaria parasites, Plasmodium falciparum (P. falciparum) is the most pathogenic and is frequently fatal if untreated in time [2]. Accurate malaria diagnosis together with improved public health data reporting system and health care access are essential for proper estimation of malaria statistics globally and especially in Nigeria.

Nigeria and the Democratic Republic of Congo have been tagged hyperendemic for malaria as both countries accounted for 40% of the more than 80% of morbidity resulting from malaria in 18 African countries in 2012 [3]. Malaria is a wide spread disease and it is of important public health concern in Nigeria because of its impact on children and maternal health [4]. It has been established as the cause for 11% maternal deaths, 60% of outpatient visits and 30% of hospitalizations in the country are malaria related cases [5]. In addition, malaria is said to kill one African whether child or adult

every 15 seconds and roughly 300,000 Nigerian children annually [6]. People who live below the poverty line, children under five years of age (22% of population) and pregnant women (20% of population) are the most vulnerable to the disease even where some degree of acquired immunity in areas of intense transmission for most adult population is offered [7].

The current approach of the WHO to control malaria in sub-Saharan Africa is a combination of vector control, in the form of Long Lasting Insectide Nets (LLINs), indoor residual spraying with insecticides (IRS), and the distribution of Artmesinin Combination Therapy (ACT) drugs for treatment [8]. Insecticide-treated nets (ITNs) have been shown to be highly efficient at reducing malaria on a community level in urban Ghana [9]; other interventions, such as larviciding and removal of vector breeding sites, are appropriate in both urban and peri-urban settings. Improved housing, for instance, by using corrugated iron instead of thatched roofing reduces entry points for mosquitoes and is appropriate in less affluent urban settings [10].

Traditional practice for outpatients has been to treat presumptively for malaria based on a history of fever but, a significant proportion of those treated may not have parasites (over 50% in many settings) and hence waste a considerable amount of drugs [11]. This old clinical based practice is still relevant today especially, in infants where time spent on getting a confirmatory laboratory diagnosis could lead to increased fatality. Alternatively, the mainstay of malaria diagnosis has been the microscopic examination of blood, utilizing blood films [12]. Although blood is the sample most frequently used to make a diagnosis, both saliva and urine have been investigated as alternative, less invasive specimens [13]. More recently, modern techniques utilizing antigen tests or polymerase chain reaction have been discovered, though these are not widely implemented in malaria endemic regions [14, 15].

The malaria burden faced by African countries continues to be a challenge for national governments with Nigeria inclusive. The fact that a vaccine to cure malaria has not been developed coupled with the Increasing resistance to drugs and insecticides, the lack of capacity to implement programs effectively and low public education about malaria is only a few of the many complications that African governments must address to effectively combat malaria [16]. The losses resulting from malaria has a ripple effect on the affected nations as these consequences drive down to individuals at the community level, hence the relevance of this research.
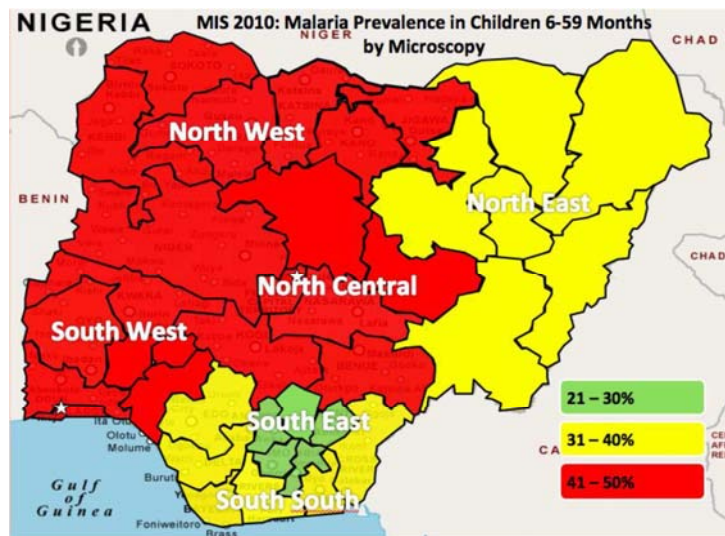
Source: WHO 2011, Nigeria Malaria fact sheet.

Figure 1. Malaria prevalence in children 6-59 months by microscopy.

2. Methodology

2.1. Study Area

Benue lies between latitudes 6°00-8°00 North of the equator and between longitudes 8°00-10°00 East of the Greenwich meridian. The average annual rainfall of Benue State is between 750mm-1300 mm annual, while the average temperature per annum is between 23°C - 35°C. The average wind speed is about 2.5m/s. Benue State has a high solar radiation with an elevation of 33,955km^2. The land surface area is dominated by grasses of various species, trees, shrubs and herbs that are usually found in this region are sparsely distributed. The population of Benue is over 4 million (census 2006). The male population is 2,164,058 while the female population is 2,055,186, a total of about 4,219,244 million.

The study was carried out in the sub-urban and urban areas of Makurdi which is situated between latitude 7°45'50"N and longitude 8°32'10"E with a population of 500,797 with Tiv, Idoma and Igede as the predominant tribes. It is located in the North Central part of Nigeria along the River Benue, a major tributary of the Niger River. The mean monthly rainfall of Makurdi ranges from 150mm to 180mm and the mean monthly temperatures ranges from 27°C to 38°C [17].

Makurdi main town comprises of Wadata, Northbank, Wurukum and High level council wards. The economic activities of the inhabitants revolves mainly around State and Federal civil service, trading, menial (unskilled) jobs, fishing and farming and other professional services. The town is divided by the river Benue into the North and South banks, which are connected by two bridges: the railway bridge, which was built in 1932, and the new dual carriage bridge built in 1978. The North bank area of the town houses among other establishments, the Federal University of Agriculture, the Nigerian Army School of Military Engineering (NASME), the headquarters of the 72 Airborne Battalion and the State Headquarters of the Department of Customs and Excise. The southern part of the town is made up of several wards, including Central Ward, Old GRA, Ankpa Ward, Wadata Ward, High Level, Wurukum (Low Level), New GRA etc. Important establishments and offices located here include the Government House, The Benue State Secretariat, The Federal Secretariat, Banks, Police Headquarters, Nigeria Prisons Service, Aper Aku Township Stadium, Nigeria Air force Base, the Makurdi Modern market, the Federal Medical Centre, Nigeria Railway Station, Radio Benue, Nigeria Television authority (NTA), Central Post Office, Benue Hotels, Benue State University etc.
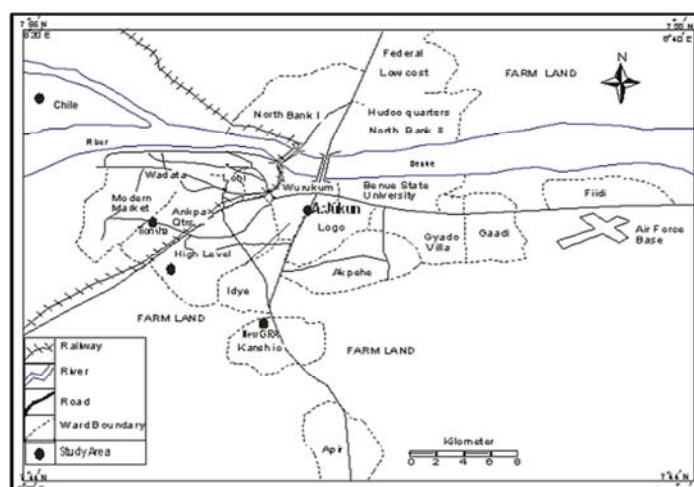
The research work was carried out in five selected areas of interest in Makurdi Local Government Area. These include:
- Angwan Jukun
- Chile Island
- High level
- New GRA
- Tionsha
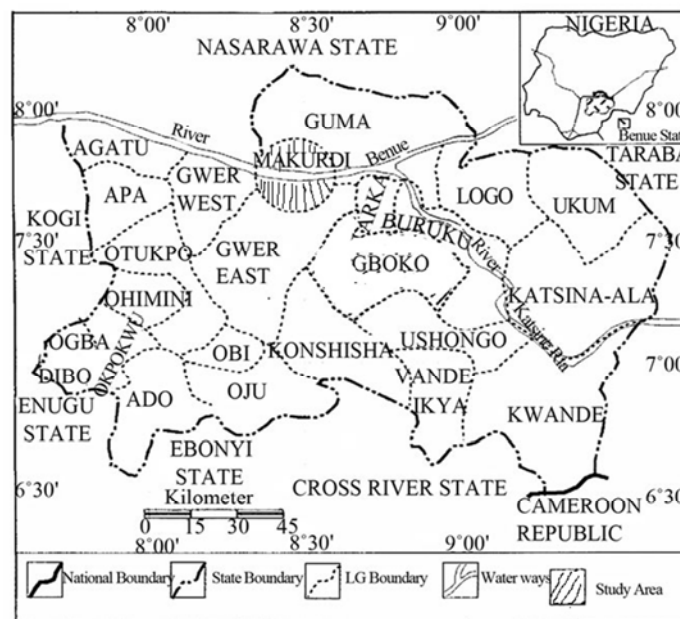
Source: Google map

Figure 2. Map of Benue State showing Makurdi.

Source: Google map

Figure 3. Map of Makurdi, showing study areas.

2.2. Sample Size Estimation and Study Design

The study was basically community based and study unit was the head of the household or their representative. In this cross sectional survey, a sample size of 328 was used in the study following guidelines for sample size calculation by Roasoft ®. Therefore, the sample size was determined based on the following considerations:

i.   Concerns related to cost was considered in determining the sample size.
ii. In the absence of accurate previous prevalence data on the population under study, calculation was made assuming 50 % of the households to have at least one perceived malaria patient over the past one year.
iii. A 95 % confidence interval was assumed.
iv. A margin of error of 0.5 was also assumed.

2.3. Ethical Considerations

This research was reviewed and approved by the Ethical Clearance Committee of the Department of Biological Sciences, Benue State University Makurdi. Written and verbal informed consent was obtained from all respondents who participated after explaining the purpose and objectives of the study in the English and local languages. Participation in the study was voluntary and the researcher ensured confidentiality of the information both during and after data collection. The respondents were informed about their rights not to participate, not to answer any question or all of the questions. Any respondent that tested positive to malaria was administered with appropriate Anti-malaria according to the national guidelines at the time of the study.

2.4. Laboratory Analysis

About 2.5 millimeters of intravenous blood samples (328 samples) were collected into well-labelled Ethylene Diamine Tetracetate (EDTA) containing bottles by well trained laboratory technicians and a medical doctor, from interested members of households in the study area.

2.5. Diagnosis

Diagnosis was done using two methods:

2.5.1. Rapid Assessment Method
This was carried out according to manufacturer's instruction with the aid of Care Start ™ malaria rapid diagnostic kit (manufactured by Access Bio, Inc USA) and purchased at Modiccum laboratory equipment stores, Beach Road, Jos- Plateau State.

2.5.2. Principle
The malaria *Plasmodium falciparum* rapid test device (whole blood) is a qualitative, membrane based immunoassay for the detection of *Plasmodium falciparum* antigen in whole blood. The membrane is pre-coated with anti-HRP2 antibody. During testing, the whole blood specimen reacts with the dye conjugate, which has been pre-coated in the test strip. The mixture then migrates upward on the membrane chromatographically by capillary action and reacts with anti-HRP2 antibody on the membrane on the test line. If the specimen contains HRP2, a coloured line will appear in the test region. The absence of the coloured line in the test region indicates that the specimen does not contain HRP2. To serve as a procedure control, a coloured line will always appear in the control region indicating that proper volume of specimen has been added and membrane wicking has coloured.

2.5.3. Rapid Assessment Procedure
Valid packs of Carestart ™ malaria HRP2 (*Pf*) kit for detection of *Plasmodium falciparum* containing; alcohol swab, lancet, plastic pipette, buffer and 25 test kits each was used. Immediately before use, the moisture proof pouch was opened to extract the cassette and other components. The cassette was placed horizontally on the working surface. 50µl of veinous blood was dropped in the sample well marked 'S' and 2 drops of clearing buffer was dispensed into the buffer well marked 'A'. Results were read and recorded after 20 minutes.

2.5.4. Interpretation of Results
The test was positive if 2 coloured lines appeared on the result window, 1 on the control (C) region and the other on the test (T) region. A negative test was indicated by the appearance of only 1 coloured line on the control (C) region and none on the test region. An invalid test was indicated by the non-appearance of a coloured line on the control region with or without a coloured line on the test region (figure 9 a and b).

2.6. Microscopy

Thick blood films were made and processed on greese-free microscope slides from the blood samples collected. The slides were stained using Romano sky dyes (Fields stain A and B) and viewed microscopically using the x100 magnification objective.

2.6.1. Preparation and Staining of Thick Films
About 50µl of blood was dropped in the middle of each greese-free frosted end slide using a plastic pipette. The base of the pipette was used to spread the blood to make a thick smear. The films were labeled at the frosted end and placed on a flat surface to air-dry [18]. This is as shown in Figure 8a and 8b.

2.6.2. Staining Procedure for Thick Films
- Already prepared Field's stains A and B were filtered and dispensed into glassware beakers and labeled appropriately.
- A beaker containing water was placed beside each beaker containing Field's stains A and B.
- Holding the slides with the dried thick film facing downwards, the films were dipped into Field's stain A for 5 seconds, excess stain was drained by gently tapping the edge of the slide against the beaker.
- The stained slides were washed gently for 2 seconds in clean water and excess water was drained off using a

sterile cotton wool to drain the edges and back of the slide.

- The slides were further dipped in Field's stain B for 2 seconds and washed gently in clean water for 2 seconds.
- Excess water was drained from the slides using a sterile cotton wool and the slides were placed vertically to air-dry.
- The stained slides were then viewed under the microscope using the oil immersion (x100) objective lens [18].
- Presence of ring forms and trophozoites of Plasmodium indicated positive results. A blood smear was considered negative if no parasite is seen after 10 minutes of search or examination under x100 high power fields of the microscope.

2.6.3. Interpretation of Result

Malaria parasitemia (MP), was defined as the presence of any asexual (or other forms) of malaria parasites on a thick blood smear.

2.7. Questionnaire Administration

A structured questionnaire was administered to each participant in the study areas. Data were collected on demographics of participants such as the literacy level, occupation, sex, age, marital status, religion e.t.c. The questionnaire was administered to different groups of people that were found in the study area. The help of interpreters was sought to translate the questionnaires in the various local languages of the respondents.

2.8. Statistical Analysis

The obtained results from the questionnaires and blood samples were analyzed using Statistical Package for Social Sciences (SPSS) version 20. The data generated from this study were presented using descriptive statistics. Chi-square was used to obtain level of significance (p<0.05). The sensitivity, specificity, and predictive values of each of the two test methods were calculated by comparing with a composite reference gold standard generated from the two methods. The composite reference method was defined as a method that is positive for malaria parasites by all the two methods (TFM and RDT) and also negative for malaria parasites by all the two methods. This gives the method 100% hypothetical sensitivity, specificity, and positive and negative predictive values (PPV and NPV). The sensitivity, specificity, and predictive values of each of the methods were then calculated using the formulae (1-4):

$$\text{Sensitivity (\%)} = \frac{TP}{TP + FN} \text{ X } 100 \qquad (1)$$

$$\text{Specificity (\%)} = \frac{TN}{TN + FP} \text{ X } 100 \qquad (2)$$

$$\text{PPV (\%)} = \frac{TP}{TP + FP} \text{ X } 100 \qquad (3)$$

$$\text{NPV (\%)} = \frac{TN}{TN + FN} \text{ X } 100 \qquad (4)$$

Where TP = true positive, FP = false positive, TN = true negative, and FN = false negative. Sensitivity was defined as the probability that a truly infected individual will test positive and specificity as the probability that a truly uninfected individual will test negative.

3. Results

3.1. Prevalence of Malaria in Relation to Sex of Respondents

Figure 4 shows the prevalence of malaria in respect to sex. Of the 328 participants examined, the overall prevalence of malaria was 32 (9.8%) by RDT and 164 (50.0%) by microscopy. The highest prevalence was recorded among females 20(11.2%) and 92 (51.7%) by RDT and microscopy. Males had a lower prevalence of 12(8.0%) and 72 (48.0%) also in order of RDT and microscopy. Statistical analysis by chi-square revealed that malaria prevalence by gender is statistically insignificant (P >0.05).

3.2. Prevalence of Malaria in Relation to Occupation

The prevalence of malaria in relation to occupation is captured in Table 1. Going by the RDT method, students 14(14.3%) scored the highest prevalence, this was closely followed by others 9(13.6%) while the least infection was recorded among traders 0(0.0%). Microscopy reported highest prevalence among others 37 (56.1%) closely followed by students 54 (55.1%) while the least prevalence was reported in farmers 13(35.1%). The prevalence of malaria in respect o occupation is statistically insignificant (P>0.05).

3.3. Prevalence of Malaria According to Educational Status

The prevalence of malaria in respect to educational status of respondents is presented in figure 5. Malaria infection was prevalent in all groups with the highest prevalence recorded in those with no formal education; 8(14.3%) and 34 (60.7%) by RDT and Microscopy. This was followed by those with primary education 12(13.6%) by RDT and 57(53.8%) among those with tertiary education, by microscopy. The least prevalence was reported in those with secondary education 5(6.4%) and 30(38.5%) (RDT and Microscopy). There was no significant difference (P>0.05) in Malaria infection and level education.

3.4. Prevalence of Malaria with Respect to Location

Table 2 shows the prevalence of malaria in respect to location of respondents. Of the five locations sampled, results from RDT showed that the highest prevalence was recorded in New GRA 15(20.3%), followed closely by Chile 12(19.0%) while high level had no infection 0 (0.0%). Microscopically, Chile 44(69.8%) scored the highest prevalence, followed by New GRA 42(56.8) and Tionsha 27(48.2%). The least infection was recorded in Angwan

Jukum 24(34.3%). for both methods, malaria infection was statistically significant to location (P=0.000).

3.5. Prevalence of Malaria in Relation to Age of Respondents

Figure 6 illustrates the level of malaria infection with respect to age group. Using RDT, the highest prevalence 4(19.0%) was recorded within age group 11-15 years, this was closely followed by age group 6-10 years with prevalence rate of 7(18.9%). Low prevalence 1(5.0%) and 5 (5.3%) was seen amongst age groups 16-20 years and 21-30 years, respectively.

Going by microscopy, the highest prevalence 13(61.9%) was recorded among age group 11-15 years, closely followed by age group 1- 5 years with 16 (61.5%) while the least prevalence 2(33.3%) was recorded among age 50 years and above. For both methods, there was no statistical significance in the prevalence of malaria with respect to age of respondents (P> 0.05).

3.6. Comparison between Care Start TM RDT and Microscopy

Taking the light microscope as a gold standard for malaria diagnosis, the sensitivity and specificity of Care Start TM RDT was found to be 16.5% and 97.0% respectively. The Positive Predictive Value (PPV) and Negative Predictive Value (NPV) were found to be 84.4% and 53.7%. (Table 3)
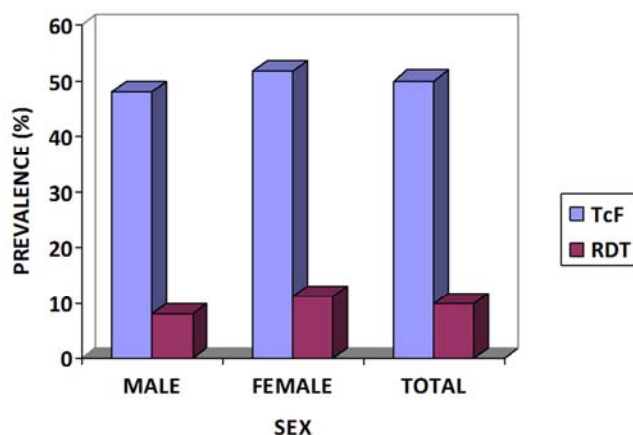

Figure 4. Prevalence of Malaria in relation to sex.

Table 1. Prevalence of Malaria in Relation to Occupation.

Occupational Status	No. Examined	No. Infected RDT	TcF
Civil Servant	69	5(7.2)	30(43.5)
Farming	37	2(5.4)	13(35.1)
Trader	42	0(0.0)	23(54.8)
Fishing	16	2(12.5)	7(43.8)
Student	98	14(14.3)	54(55.1)
Others	66	9(13.6)	37(56.1)
Total	328	32 (9.8)	164 (50.0)
χ^2		9.397	7.065
Df		5	5
P- value		0.095	0.216

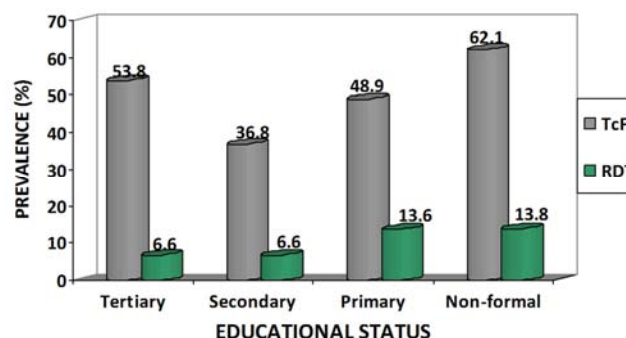RDT (Rapid Diagnostic Test), TcF (Thick film)

Figure 5. Prevalence of Malaria in Relation to Educational Status.

Table 2. Prevalence of Malaria in Relation to Location.

Location	No. Examined	No. Infected (%) RDT	TcF
Agwan Jukum	70	1(1.4)	24(34.3)
Chile Island	63	12(19.0)	44(69.8)
High level	65	0(0.0)	27(41.5)
New GRA	74	15(20.3)	42(56.8)
Tionsha	56	4(7.1)	27(48.2)
Total	328	32(9.8)	164(50.0)
χ^2		28.444	20.119
Df		4	4
P-value		0.000**	0.000**

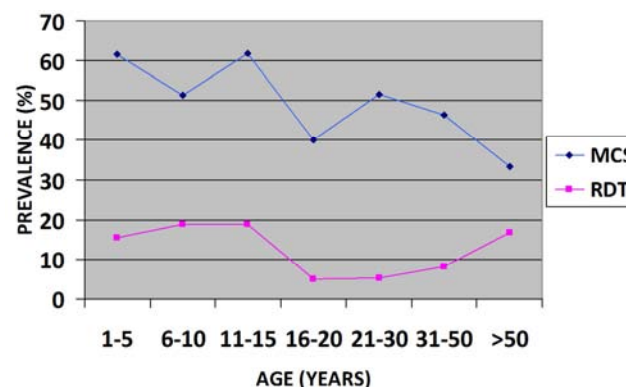** highly significant, RDT (Rapid Diagnostic Test), TcF (Thick film)

Figure 6. Prevalence of Malaria in Relation to Age.

Table 3. Comparison between Care Start TM RDT and Microscopy.

Microscopy	RDT		Total
	Positive (%)	Negative (%)	
Positive	TP 27(8.2)	FP 5(1.5)	32
Negative	FN 137(41.8)	TN 159(48.5)	296
Total	164	164	328

TP= True positive, FP= False positive, FN=false negative, TN= True negative
Sensitivity=16.5%, Specificity= 97.0%, Positive predictive value (PPV) =84.4%, Negative predictive value (NPV)=53.7%

4. Discussion

Malaria is prevalent in Makurdi, Benue State and it's of immense public health importance. Overall prevalence by Care Start TM Rapid Diagnostic Test (RDT) was 9.8% while microscopy which is regarded as a gold standard reported a

prevalence of 50.0%. This record is same with another research [19] and similar to reports (49.0%) from study among pregnant who women attended Federal medical Centre and Bazz laboratory in Makurdi [20]. Other studies also showed a lower prevalence (42.4%) from Otukpo and a higher (60.9%) prevalence in Makurdi, both in Benue state [21, 22]. Lower prevalence 17% and 21 % has been recorded in other parts of the country [23, 24]. The > 40% annual rate reported in Nigeria suggests that the prevalence of malaria in Nigeria varies with location [25]. The climatic factors, environmental and behavioural pattern of people in Makurdi which promote breeding and the susceptibility of the people to vector bites could be responsible for the prevalence rate recorded in this study.

Malaria prevalence among sexes was not statistically significant (P>0.05). Malaria parasitaemia was slightly higher among females than males going by both RDT and microscopy. Findings are similar to results presented from other parts of Nigeria [26-30] (Kalu *et al.,* 2012, Onyido *et al.,* 2011, Nwuzo *et al.,* 2009, Nebe *et al.,* 2007, Ezeanya 1998,). Contrasting findings were recorded among Cameroonian pupils where males (43%) has higher prevalence compared to females (27%) [31]. Also, reports from the eastern Nigeria are different from findings in this study [32, 33]. The higher infection rates recorded in females may be due to physiological differences between females and males (Ovulation, pregnancy and child birth) which tend to lower the female immunity, thus predisposing them to malaria infection and other diseases.

Results show high prevalence of malaria (61.9% and 19.0%) among age group 11-15 years by microscopy and RDT least (33.3% by microscopy) among individuals that were 50 years and above (5.0% by RDT) among age group 16-20 years. Similar findings were recorded by other researchers [26, 34]. The high rate of infection among these individuals may be attributed to lack of protection against mosquito bites or lack of knowledge of malaria transmission or both. The equally high rate of infection (61.5%) recorded among age group 0-5years by microscopy was similarly reported in Indonesia and also among primary health facilities attendees in Ogun state, Nigeria [32, 35]. This level of prevalence may be due to lack of transferred maternal immunity or infection acquired through the mother as in the case of congenital malaria.

Further stratification of prevalence according to educational status gave a statistically insignificant result (P>0.05). The highest prevalence (14.3% - RDT and 60.7 % - microscopy) was found among those with no formal education. This finding corroborates other researchers' findings [32]. This prevalence could be tied to ignorance and poverty. It has been noted that in addition to abundant mosquito breeding sites in the environment, ignorance, poverty, unsanitary conditions, poor behavioural attitudes and inadequately planned socioeconomic projects tended to increase malaria transmission in rural areas [36]. From this study, it shows that education invariably affects people's perceptions about causes of certain diseases of which malaria

is not an exception. Although previous reports indicate that increasing literacy level has no direct correlation with best malaria practices [37, 38], and that increasing literacy level will only serve as a protective factor against malaria morbidity [39, 40].

Prevalence of malaria parasitaemia and occupation was statistically insignificant (P>0.05) among different occupational groups. Microscopy showed the highest prevalence among the group 'others', followed by students (56.1% and 55.1%). This contrasts the findings reported with highest prevalence (94.34% and 93.75%) among urban and rural traders in Eastern Nigeria [26]. Results from RDT showed high prevalence among students (14.3%) followed by others (13.6%) while no case was reported among traders. Going by both methods, students were more open to malaria. This suggests inadequate protection resulting in greater exposure to mosquito bites. People within the age of schooling mostly range from children to youths who are mostly likely to be involved in activities that enhance their exposure to mosquito bites.

Comparing the conventionally accepted light microscopy of peripheral blood slides with the Care Start TM malaria HRP2 Rapid Diagnostic Test (RDT), more infection were detected by blood slide microscopy, 164 (50%) than by RDT, 32 (9.8%). The findings in this current study are similar to other reports [41-43].

Considering the light microscopy as a standard, this study revealed a sensitivity of 16.5% and specificity of 97.0%. RDT had a high Positive predictive value (PPV) of 84.4% meaning respondents stand the chance of being correctly diagnosed as positive for malaria, avoiding unnecessary treatment. The moderate Negative Predictive Value (NPV) of 53.7% indicates that RDT was not strongly reliable in ruling out the chance of malaria. In another research, different findings were reported from a comparison between Care Start TM malaria HRP2 Rapid Diagnostic Test (RDT) and peripheral blood slides microscopy using samples for patients attending Hajiya Gambo Sawaba General hospital in Zaria. In the study RDT showed sensitivity (78.4%), Specificity (97.6%), PPV (97.3%) and NPV (80.1%) [43]. The marked difference between the various findings can be attributed to the fact that it is scientifically expected for the RDT to be more accurate in detecting clinical episodes of malaria since the concentration of the parasites in the blood will be higher at such level which is not the case among asymptomatic community respondents in this study.

5. Conclusion

Malaria is present in Makurdi, Nigeria. The prevalence of the infection in all five locations 164 (50%) supports the endemicity of malaria among members of households in Makurdi and this poses a public health challenge to the inhabitants and economy of Makurdi. This research also revealed a high prevalence in respect to sex, age, educational and occupational status. The low sensitivity of the Care Start [TM] RDT compared to light microscopy which was observed

in this study makes it unsuitable as a choice tool for diagnosis of asymptomatic malaria in patients and since it is also unable to detect parasites but rather antigens, results may therefore reflect recent and non-current parasitaemia.

Appendix

Source: Researcher's field work

Figure 8b. Air-dried thick films.

Source: Researcher's field work

Figure 7. Sample collection from respondents.

Source: Researcher's field work

Figure 9a. Sample of negative RDT result

Source: Researcher's field work
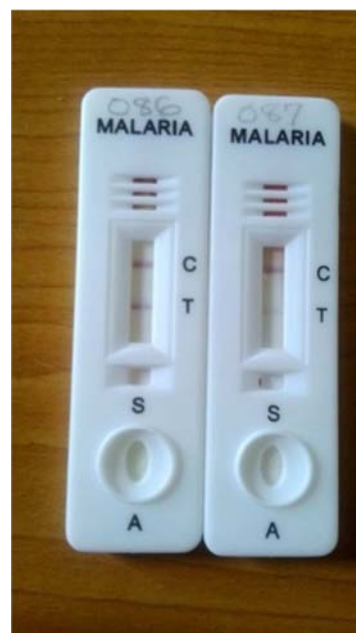
Figure 8a. On-spot preparation of thick films.

Source: Researcher's laboratory work.

Figure 9b. Samples of positive results obtained by RDT.

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