International Journal of TROPICAL DISEASE & Health
21(2): 1-10, 2017; Article no.IJTDH.30277
ISSN: 2278–1005, NLM ID: 101632866

SCIENCEDOMAIN *international*
www.sciencedomain.org

Prevalence of Malaria Infection and Reliability of ACCUCARE One Step Malaria Test® for Diagnosing Malaria in People Living with Human Immunodeficiency Virus Infection in Cameroon

Lugarde Mawabo Kamgain[1,2*], Jean-Paul Assam-Assam[2,3], Loick Pradel Kojom Foko[4] and Henri Lucien Kamga Fouamno[2,5]

[1]District Hospital of Deido, Douala, Cameroon.
[2]Institut International d'Afrique Centrale (IIAC), P.O.Box 293, Moundou, Chad.
[3]Department of Biochemistry, Faculty of Science, University of Douala, P.O.Box 24157, Douala, Cameroon.
[4]Department of Animal Biology, Faculty of Science, University of Douala, P.O.Box 24157, Douala, Cameroon.
[5]Department of Medical Laboratory Science, Faculty of Health Sciences, University of Bamenda, P.O.Box 39, Bambili, Cameroon.

Authors' contributions

This work was carried out in collaboration between all authors. Author LMK designed the study, wrote the protocol and wrote the first draft of the manuscript. Author JPAA designed the study, supervised the work, read and corrected the manuscript. Author LPKF performed statistical analyses, read and corrected the manuscript. Author HLKF designed the study, wrote the protocol and supervised the work at all stages. All authors read and approved the final manuscript.

Article Information

DOI: 10.9734/IJTDH/2017/30277
Editor(s):
(1) Shankar Srinivasan, Department of Health Informatics, University of Medicine & Dentistry of New Jersey, USA.
(2) Nicolas Padilla-Raygoza, Department of Nursing and Obstetrics, Division of Health Sciences and Engineering, Campus Celaya Salvatierra, Mexico.
Reviewers:
(1) Dawit Getnet Ayele, The Johns Hopkins University, USA.
(2) Ikegbunam Moses Nkechukwu, Nnamdi Azikiwe University, Nigeria.
(3) Okolie Vitus Ezike, Nnamdi Azikiwe University Teaching Hospital, Nigeria.
(4) Florencia del Puerto, National University of Asuncion, Paraguay.
Complete Peer review History: http://www.sciencedomain.org/review-history/17634

Original Research Article

Received 29th October 2016
Accepted 23rd January 2017
Published 27th January 2017

**Corresponding author: Email: lugyk@yahoo.fr;*

ABSTRACT

Aims: This study aimed at determining the malaria prevalence and appraising the diagnostic performances of a rapid diagnostic test (RDT), namely ACCUCARE one step Malaria Test®, for malaria in people living with human immunodeficiency virus infection (PLWHIV).
Study design: This study was a cross-sectional study.
Place and Duration of Study: The study was carried out at the District hospital of Deido in Douala, Cameroon between August 2015 and March 2016.
Methodology: A total of 723 patients were included in the study. Malaria parasites were detected using Giemsa-stained blood films and RDT. The reliability of the RDT was evaluated by calculating the sensitivity (Se), specificity (Sp), positive predictive value (PPV), negative predictive value (NPV), accuracy and Kappa index (κ). Results were analyzed and compared using light Giemsa-stained blood films as gold standard. HIV infection was confirmed using methods based on immunochromatography and ELISA. CD4 lymphocytes count was determined by flow cytometry to depict the immune status.
Results: Overall, Plasmodia were found in 121 (16.7%; 95%CI: 14.2%-19.6%) and 91 (12.6%; 95%CI: 10.3%-15.3%) using light microscopy and RDT respectively. The sensitivity, specificity, PPV and NPV of the RDT were 75.2% (95%CI: 66.8%-82.1%), 100% (95%CI: 99.4%-100%), 100% (95% CI: 95.9% -100%) and 95.3% (95% CI: 93.3% - 96.7%) respectively. The agreement between both methods was excellent (κ = 0.835; P < .0001).
Conclusion: The study showed a good performance of the RDT in terms of specificity, PPV, NPV, accuracy and agreement. This test might represent a good alternative to the standard method for diagnosis of malaria in PLVIH.

Keywords: Malaria; rapid diagnostic test; diagnosis performance; HIV-infected people; Cameroon.

1. INTRODUCTION

Malaria is still a major public health concern throughout the world and especially in Africa. According to the latest WHO estimates, this disease was responsible for 214 million and 438,000 cases and deaths respectively in 2015. The burden is more important in Sub-Saharan Africa countries which account for 88% and 90% of all cases of morbidity and mortality [1]. Sub-Saharan Africa (SSA) is also the region the most burdened by the human immunodeficiency virus (HIV) infection. It is estimated that 25.8 million of people live with HIV in SSA, accounting for about 70% of the global total [2].

In Cameroon, these both diseases are causes of concern. Malaria is endemic in Cameroon with a prevalence rate of 29% [3]. In addition, malaria accounts for 40 to 50 % of medical consultations, meanwhile 30 to 47 % of hospitalizations [4]. It is the main cause of morbidity in children under five (18%), pregnant women (5%), people living with HIV (5.5%) and the poor (40%) [5]. The prevalence of HIV infection was estimated at 4.3% in 2011 over the country with the highest rates recorded in the South (7.2%), East (6.3%) and North-West (6.3%) regions [6]. Malaria and HIV are both common in SSA and share in some common risk factors including poverty. Due to their overlapping distribution, co-infections with malaria and HIV are frequent in SSA, especially in Cameroon [7]. Some authors reported the prevalence of the co-infection in Cameroon [8-10]. In 2016, Njunda et al. [10] recently recorded a prevalence of 7.3% of Cameroonians living in Yaounde.

Malaria is thought to be associated with a morbidity and mortality both increased in people living with HIV (PLWHIV). Globally, the risk of uncomplicated and severe episodes of malaria is higher in HIV patients co-infected with malaria parasites, especially in groups at risk such as pregnant women and children [11-12]. Conversely, malaria parasites increase the ability of the virus to replicate via mechanisms mainly involving an impairment of the immune response of the host [11-13].

The key to the proper management of malaria in PLWHIV is prompt and accurate diagnosis followed by appropriate treatment. Diagnosis usually relies on clinic and light microscopy in most malaria endemic areas which are often resources-constrained and remote [2-3]. Many authors reported poor performances of clinical diagnosis in terms of sensitivity and specificity [1,14]. On the other hand, light microscopy is used as a gold standard to confirm any clinical

suspicion of malaria in some healthcare facilities of these countries especially in Cameroon. This method is cheap and simple but labour intensive, time consuming, cannot detect sequestered *Plasmodium falciparum* parasites and requires well-trained personnel especially for detection of low parasite densities [15]. Overcoming these limitations is very important in order to avoid unnecessary use of artemisinin-based combination therapies (ACTs), due to overdiagnosis, as this could lead to the development of drug resistance [1]. Furthermore, a significant proportion of patients who need treatment could not promptly receive it or not at all. As a result, the risk of malaria-associated severe adverse effects and deaths might be increased in malaria infected individuals. This matter takes sense when over- and underdiagnosis fractions are mainly important in groups at risk such as children, pregnant women along with immunocompromised people as those infected with human immunodeficiency virus.

It looks critical to develop and implement new diagnosis alternative upon their evaluation in community especially in remote areas. Rapid diagnostic tests (RDTs) based on immunochromatography have been developed to address the limitations of light microscopy. RDTs are simple and easy to use with little expertise and skills required for interpretation. There are numerous malaria RDTs that are commercially available in many formats and the variation depends also on malaria antigen or antibody targeted. The commonly used RDTs target *P. falciparum* histidine-rich protein 2 (PfHRP2) and two enzymes in the parasite glycolytic pathways, namely plasmodial lactate dehydrogenase (pLDH) and aldolase [16]. Others RDTs target antibodies specific to malarial antigens by using recombinant antigens first pre-coated on the membrane strip of the RDT [16]. ACCUCARE one step Malaria Test® RDT is one of a few RDTs present in the Cameroonian market and used for diagnosing malaria infection in some health facilities of the country. Many authors have addressed the issue about the utility of RDTs in malaria endemic areas [16-18] but however, little is known in Cameroon [19-22] especially on the ACCUCARE one step Malaria Test® RDT and PLWHIV.

Thus, this study aimed to determine the prevalence of malaria infection and appraise the diagnostic performances of the ACCUCARE one step Malaria Test® RDT for malaria in HIV-infected people.

2. MATERIALS AND METHODS

2.1 Study Site

This study took place in the town of Douala (Littoral Region, Cameroon). Douala is located 3°48'N, 10°08'E, near the Atlantic coast, within the Congo-Guinean phytogeographical zone characterized by a typical equatorial climate with two rainy seasons extending from March to June and from September to November [23]. The city is 1 m above sea level and receives over 3,500 mm rainfall annually. These environment conditions are propitious for creation of breeding sites malaria vectors. Douala is a port city where many worse behaviors (liquors consumption, prostitution) significantly increase the risk of sexually-transmitted diseases (STDs) such as Human immunodeficiency virus infection. Douala is the main business city of the country and is ranked sixth (5.5%) in terms of HIV prevalence rate [6]. Participants were recruited at the district hospital of Deido which greets people coming from all parts of Cameroon owing to its strategic location and sustainable and constant supply with CD4 cells reagents and antiretroviral drugs for management of people living with HIV (PLWHIV).

2.2 Study Population

The study population consisted of individuals aged 5-49 years old of both sexes living with HIV and under highly active antiretroviral therapy. They were recruited in a convenient way and therefore a total of 723 patients were included in the study. Using the prevalence of 29.4% reported by Nkuo Akenji et al. [24], the sample size was determined using the Lorentz's formula $n = Z^2pq/d^2$ where n = the sample size required, Z = 1.96: confidence level test statistic at the desired level of significance, p = 29.4%: proportion of HIV prevalence, q = 1-p: proportion of malaria negative participants and d = accepted error willing to be committed. The minimum sample size was estimated as n = 319. Any patient who did not meet any of these abovementioned criteria were excluded from the study.

2.3 Study Design

This cross-sectional and prospective study was carried out between August 2015 and March 2016. Participants included in the study were HIV-infected, attending at the district hospital of Deido for routine control and willing to

participate. Prior to their inclusion, they were given information, education and communication on malaria and HIV infection. Thereafter, informed consent forms were signed by each participant following explanation of objectives of the study to them. Approval of parents or guardians of children was also sought. A structured questionnaire was used to collect socio-demographic, clinic and biological data. Axillary temperature was recorded using an electronic thermometer. Blood samples were collected and transported to the Laboratory of the district hospital of Deido for parasitological analyses. Investigative methods included a questionnaire approach, clinical and parasitological analyses.

2.4 Blood Collection

About five milliliters (5 mL) were collected from each participant by venipuncture into sterile plastic syringes. Blood samples were then transferred into EDTA and dry tubes for all performing hematological, parasitological and biological analyses respectively. All tubes were labeled with the patients' barcode and pathology number. Blood samples were centrifuged at 3000 rpm for 3-5 minutes and sera were stored at 2-8℃ until analysis.

2.5 Confirmation of HIV Infection

Prior to their inclusion in the study, the participants were tested for HIV infection for confirmation. HIV diagnosis was performed using Determine Alere HIV 1/2 rapid diagnosis test followed by Immunocomb HIV1/2 (Organics) accordance to the national guidelines. The former test is an immuno-chromatographic rapid test for qualitative detection of antibodies specific to HIV in human serum, plasma or whole blood. The latter is based on the enzyme-linked immunosorbent assay (ELISA) principle.

2.6 CD4 Cells Count

CD4+ T lymphocytes were counted with a flow cytometer CyFlow® (Sysmex-Partec Görlitz, Germany) according to the manufacturer's instructions. Briefly, 20 µL of phycoerythrine-conjugated monoclonal antibody to human CD4 (mAb PE MEM241, Partec GmbH, Görlizt, Germany) were slightly mixed with 20 µL of whole blood into a test tube and incubated for 15 minutes at room temperature, protected from light. Then, 800 µL of no-lyse buffer were added to the mixture. After homogenizing the content, the tube was introduced into the CyFlow Counter for automatic counting [25]. The CD4 results were grouped into as followed: < 200; 200-350; 350-500 and > 500 cells/µL.

2.7 Malaria Diagnosis

Two methods were used to diagnose malaria infection namely Giemsa-stained blood films and the ACCUCARE one step Malaria Test® RDT. Asymptomatic malaria was defined as the presence of malaria parasite with an axillary temperature of <37.5℃. Symptomatic malaria was defined as the presence of malaria parasite with an axillary temperature of ≥ 37.5℃ [26].

2.7.1 Thick blood smears

Thick blood films were performed using the protocol previously described by Cheesbrough (2004) [15]. Briefly, thick smears that were air-dried for 30 minutes, was stained with 10% Giemsa for 20 minutes. Thereafter, stained slides were allowed to air dry and stored not more than one day until microscopic examination. Microscopy was used for identification of malaria parasites by a senior. Thick blood films were considered positive when asexual forms (trophozoites and schizonts) and or gametocytes were present in the blood film. Slides were declared negative after observing at least 100 high power fields without detecting any parasites. In order to ensure quality assurance of parasitological data, thick smears-based results were classified as valid (positive or negative slides) and invalid (not read slides) as outlined in literature [26].

2.7.2 ACCUCARE one step Malaria Test®

This rapid diagnostic test (Labcare Diagnostics, India) is used to qualitatively detect antibodies of all isotopes (IgG, IgM, IgA) specific to *Plasmodium falciparum* (Pf) and *Plasmodium vivax* (Pv) simultaneously in blood samples (serum, plasma and whole blood). This consists of one control band and two test bands. The former test band, referred to as test band 1, is pre-coated with recombinant malaria Pf capture antigen (MSP, CSP) meanwhile the test band 2 with recombinant malaria Pv antigen (MSP, CSP). The recombinant malaria Pf/Pv antigen-colloid gold conjugate and serum sample moves along the membrane chromatographically to the test regions and forms a visible line outlining the presence of antibodies translating a malaria infection [27].

The diagnosis of malaria infection using this test was performed according to the manufacturer's instructions. Briefly, 10 µL of serum was added to the sample well "S". Then, 1 drop of the assay chase buffer was added. Results test were read after 10 to 20 minutes [27].

2.8 Ethical Approval and Consent

This study was carried out in conformance with the guidelines for human experimental models in clinical research as stated by the Cameroon Ministry of Public Health and the Helsinki declaration. Besides, the ethical clearance was sought close to Institutional review board (IRB) of Douala under N° CEI-UD/273/10/2015/T. The aim and objectives of the study were explained to them in the language they understood best (French or English), and their questions were answered. Only individuals who signed informed consent forms for their participation were enrolled. Participation in the study was strictly voluntary and patients were free to decline answering any question or totally withdraw if they so wished at any time. All malaria positive patients were treated on the spot accordance to the national treatment guidelines.

2.9 Statistical Analyses/Methods

All data were verified for consistency, coded, and keyed in an Excel sheet. Thereafter, statistical analyses were performed with SPPS 20.0 for Windows (SPSS, Chicago, IL, USA). Data were summarized in table as percentages with 95% confidence interval (95%CI) or mean ± standard deviation (SD) for qualitative and quantitative variables respectively where appropriate. Student's test was used to compare differences for normally-distributed variables. Chi-square test (χ^2) or Fisher's exact probability were computed to compare categorical variables. McNemar's test was used to compare malaria-based results between light microscopy and RDT. Significant levels were measured at 95% CI with significant differences recorded at $P < .05$.

After all the tests were performed the values for sensitivity, specificity, predictive values and accuracy were calculated using light microscopy as a gold standard. The variables measured were the number of true positives (TP), number of true negatives (TN), number of false positives (FP) and number of false negatives (FN). Sensitivity was calculated as TP/(TP+FN), Specificity was calculated as TN/(TN+FP), Positive predictive value (PPV) was calculated as PPV=TP/TP+FP, Negative predictive value (NPV) and accuracy were calculated as NPV=TN/TN+ FN, Accuracy = (TP+TN)/number of all tests, respectively.

The agreement between light microscopy and RDT was appraised by computing the Cohen's Kappa index [28]. Its interpretation was made based on the scale by Landis and Koch (1977) who splitted the kappa index into five categories namely very poor (< 0.00), poor (0.00-0.20), moderate (0.21-0.60), good (0.61-0.80) and excellent (≥ 0.81).

3. RESULTS

3.1 Baseline Characteristics of the Participants

A total of 723 patients were included in the study. Majority of them were females (74.3%; $P < .0001$). The mean age was 39.49 ± 11.17 years old and this variable ranged between 4 and 74. In addition, patients aged 20-49 years old and having CD4 cells count > 500 cells/µL accounted for more than two thirds (79.7%) and the third (32.9%) of the participants (Table 1). The mean CD4 count of these patients was 427 cells/µL (sd = 257).

3.2 Prevalence of Malaria

Overall, *Plasmodia* were found in 121 (16.7%; 95%CI: 14.2%-19.6%) and 91 (12.6%; 95%CI: 10.3%-15.3%) using Giemsa-stained blood films and RDT respectively. In addition, a statistically significant difference ($P < .0001$) was found using the McNemar's test. All patients infected with malaria parasites were asymptomatic and *Plasmodium falciparum* accounted for all case of infection. As presented in Table 1, malaria prevalence was higher in females (12.9%) and patients having CD4 cells count < 200 cells/µL (23.8%). On average, malaria prevalence was increasing with age of participants. Malaria prevalence was highest in above 49 years old (13.4%). Furthermore, none statistically significant differences were found (Table 1).

3.3 Diagnostic Performances of ACCUCARE One Step Malaria Test®

Out of 121 positive tested by the Giemsa-stained light microscopy, 91 have been found to be positive by the RDT while 30 were negative. On the other hand, out of 602 which were negative

Table 1. Baseline characteristics of the participants and their relation to malaria infection

Variables	Levels	Total		Positive[§] (%)	P-value
		n	%		
Gender	Female	541	74.3	70 (12.9)	0.6220
	Male	182	25.7	21 (11.5)	
Age (years)	≤ 19	13	1.8	0 (0.0)	0.3750
	[20 - 50[	576	79.7	73 (12.7)	
	≥ 50	134	18.5	18 (13.4)	
CD4 count (cells/µL)	< 200	122	16.9	29 (23.8)	0.0991
	200 - 350	192	26.6	32 (20.0)	
	350 - 500	171	23.6	22 (12.9)	
	> 500	238	32.9	38 (15.9)	

Data are presented as frequency (percentage). Independent chi square was used to compare proportions. P-value < 0.05 was considered as significant. §: malaria prevalence was calculated based on the gold standard (Giemsa-stained blood films)

by Giemsa stained light microscopy, none were positive by the RDT (Table 2). Thus, the sensitivity and specificity of RDT was 75.2% and 100% respectively as summarized in Table 3. Besides, the agreement between both methods was excellent (κ = 0.835; *P* < .0001). This matter was a good concordance between the methods because of an accuracy of 95.8% (Table 3).

4. DISCUSSION

Reliable and convenient diagnostic method is critical to fight against malaria infection. Clinic- and microscopy-based diagnoses are commonly used in malaria endemic areas which are often resource-constrained. Unfortunately, several concerns emerged about their cost-effectiveness to efficiently control malaria due to their intrinsic limitations. Clinic diagnosis is low sensitive and specific mainly in high transmission areas. Microscopy is time-consuming, labour-intensive and depends on quality of reagents and competence of the microscopist. Thus, rapid diagnostic tests (RDTs) may represent an interesting and reliable alternative especially in remote areas where electricity is missing. This study aimed at appraising the performances of a RDT for diagnosis of malaria in people living with HIV infection.

4.1 Prevalence of Malaria Infection

The malaria prevalence by light microscopy was 16.7% in the participants. This value is lower than that found by many reports [29-31]. These authors found 31.76%, 24.0% and 74.3% respectively using the light microscopy as well. Conversely, our value is higher than the 11.75% found by Tay et al. [32] in Ghana. Differences in sample size, study design, study period, study area along with genetic background and behavioral patterns of individuals can explained the discrepancies observed between these study and ours.

4.2 Association between Malaria Infection and CD4 Cells Count

In this study, we found negative, although not significant, between malaria prevalence and CD4 lymphocytes counts. However, our finding is quite consistent with Van Geertruyden et al. [33] who outlined an slower increasing in CD4 cells counts in malaria-infected HIV patients than their malaria uninfected counterparts after successful antimalarial treatment. Furthermore, Chavale et al. [34] also outlined the $CD4^+$ T cell counts were significantly lower in the *Pf*/HIV group than in the HIV/AIDS only or malaria only patients in Mozambique. Thus, HIV and malaria diseases are both controlled by immunity, and decreasing immune status as found in HIV patients will cause an increase in malaria severity. Besides, this matter outlines the need for diagnosing malaria infection in PLHIV during follow up.

4.3 Diagnostic Performances of the ACCUCARE One Step Malaria Test®

A sensitivity of 75.2 % (95%CI: 66.8%-82.1%) was recorded in this study. This value is lower than that of Suchita et al. [35] and Khatib et al. [36] who found 95.24% and 92.95% in India using the same RDT. Ali et al. [21] found values of specificities greater than 90%, on average, by appraising a set of 22 RDTs available in the Cameroon market. These discrepancies may be explained by the spatial and time variability of confounders such as age structure of population or coverage rate of malaria-related preventive methods. Ayele and colleagues [37-38] have

Table 2. ACCUCARE one step malaria Test® results compared to Giemsa stained light microscope

		RDT results		
		Positive	**Negative**	**Total**
Giemsa stained light microcopy results	Positive	91 (TP)	30 (FP)	121
	Negative	0 (FN)	602 (TN)	602
	Total	**91**	**632**	**723**

**Data are presented as frequency. RDT = Rapid diagnosis test, TP = True positive, FP = False positive, FN = False negative, TN = True negative*

Table 3. Diagnostic performance of ACCUCARE one step malaria Test® using light microscopy as gold standard

Performance	Values
Sensitivity, % (95%CI)	75.2 (66.8-82.1)
Specificity, % (95%CI)	100% (99.4-100)
Positive predictive value, % (95%CI)	100% (95.9-100)
Negative predictive value, % (95%CI)	95.3 (93.3-96.7)
Accuracy, % (95%CI)	95.8 (94.1-97.1)
Kappa index, (standard error, *P*)	0.835 (0.029; *P* < .0001)

**95%CI: Confidence interval; P < .05 are considered significant*

addressed this issue in details. Our finding outlines the fact that the ACCUCARE one step Malaria Test® RDT was not in line with the gold standard in 25% of patients. Thus, a significant fraction of population would not have received treatment if the test was used alone for malaria diagnosis. As a result, starting on malaria-related treatment will have been delayed in these patients and consequently increasing the risk of severe malaria.

Specificity of 100% was recorded and therefore no falsely-positive case was reported in this study. This is in line with Suchita et al. [35] but slightly different from previous studies [20-22, 36]. For instance, Sayang et al. [20] found a specificity of 82.2% using the Diaspot® RDT in outpatients in a peripheral urban health facility in Cameroon. However, these findings witness all the highly specific nature of the ACCUCARE one step Malaria Test® to exclude any case of malaria in malaria-related asymptomatic PLWHIV. It is particularly important in the context of reducing drug exposition to patients who do not need antimalarial drugs. Thus, overtreatment of patients could efficiently mitigated and consequently the drug pressure which accounts mainly for appearance and spreading of drug-resistant malaria parasites. This would be helpful to extend the use of artemisinin-based therapeutic regimens used for treating malaria in most of endemic countries [1]. Besides, it would be interesting to investigate the clinical utility of this RDT in malaria-related febrile HIV patients in further studies in our context.

Positive and negative predictive values of the ACCUCARE one step Malaria Test® RDT were high (100% and 95.3% respectively). Our results are higher than those found by previous authors in Cameroon [20,22]. Positive and negative predictive values estimates the probability for a patient of being declared positive (negative) with the gold standard given this patient was positive (negative) with the RDT respectively. This RDT might therefore be a good adjunct to light microscopy in certain conditions for diagnostic of PLWHIV. Indeed, given these characteristics, this might be particularly helpful are in remote malaria endemic areas along with hard-to-reach population where health facilities and microscopes are not often available. Besides, electricity source is also often missing which is needed to perform microscopy-based malaria diagnosis. Thus, utilization of RDTs, which does not request electricity, of high diagnostic performances would be particularly cost-effective for control malaria in such areas. This assumption is worth investigating in the next coming years in our context.

Unfortunately, parasite density has not been measured and therefore represents an important limitation in this study. Many authors previously outlined the performances of RDTs vary as a function of parasite density [20,22]. For instance, Tahar and colleagues showed the sensitivity of

the three RDTs they evaluated was between 30.8% and 66.7% at parasite densities (DP) less than 500 parasites per µL. This switched to between 98.9% and 100% at DP more than 5000 parasites per µL [22]. This is particularly important in terms of clinical utility of RDTs in epidemiologically different settings especially in low endemic where the performances of RDTs are usually better [39] as in these areas the chances of being infected with high parasite densities are higher [40]. To be noted, Douala might be referred to as highly malaria endemic area based on recent entomological results of Antonio-Nkondjio and colleagues [23]. Thus, the relatively good diagnostic performances of the RDT tested in this study should be taken with caution given these were evaluated without taking into account parasite density and in a fraction of population at higher risk of malaria. In these individuals, it is likely to record parasite densities higher than in general population. Further studies are needed to confirm these first findings of the study as well as investigate the performances of RDTs commercially available in our context.

5. CONCLUSION

The study showed a good performance of the RDT in term of specificity, PPV, NPV, accuracy and agreement. Thus, this test might represent a good alternative to the standard method for diagnosis of malaria parasites in asymptomatic carriers co-infected with HIV although parasite density was not measured in this study. Further studies are needed to confirm its long term performances along with others assumptions raised in this study.

ACKNOWLEDGEMENTS

The authors are grateful to all patients who participated in the study as well as the staff of the district hospital of Deido for their technical assistance.

DISCLAIMER

The findings and conclusions in this paper are those of the authors and do not necessarily represent the views of any of their affiliated Research Institutions. Furthermore, mention of any company name or product does not constitute endorsement by the authors. None financial support has been allocated for this study.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. World Health Organization. WHO global malaria programme: World malaria report. Geneva: World Health Organization. 2015;280.
2. UNAIDS. Fact sheet; 2015. Available:http://www.unaids.org/sites/default/files/media_asset/20150901_FactSheet_2015_en.pdf (Accessed 30 Nov 2015)
3. Mangham LJ, Cundill B, Achonduh OA, Ambebila JN, Lele AK, Metoh TN, Ndive SN, Ndong IC, Nguela RL, Nji AM, Orang-Ojong B, Wiseman V, Pamen-Ngako J, Mbacham WF. Malaria prevalence and treatment of febrile patients at health facilities and medicine retailers in Cameroon. Trop Med Int Health. 2012;17: 330–42.
4. Ministry of Public Health: National malaria control programme report. Republic of Cameroon; 2008. Available:http://www.who.int/alliance-hpsr/projects/alliancehpsr_policybriefscalingupmalariacameroon.pdf (Accessed 15 August 2015)
5. Bigoga JD, Manga L, Titanji VP, Coetzee M, Leke RG. Malaria vectors and transmission dynamics in coastal south-western Cameroon. Malar J. 2007;6:5.
6. Enquête démographique sur la santé (EDS-MICS). Rapport Préliminaire sur la prévalence du VIH. Enquête Démographique et de Santé et à Indicateurs Multiples 2011. Yaoundé ; 2012. French.
7. Hochman S, Kim K. The Impact of HIV and Malaria Coinfection: What Is Known and Suggested Venues for Further Study. Interdi Pers Infect Dis. 2009;1-8.
8. Nkuo-Akenji T, Tevoufouet EM, Nzang F, Ngufor N, Fon E. High prevalence of HIV and malaria co-infection in urban Douala, Cameroon. Afr J AIDS Res. 2008;7(2): 229–35.
9. Kombi HK, Njoh DT, Ndamukong KJN. Malaria in HIV/AIDS patients at different CD4+ T cell levels in Limbe, Cameroon. J Bacteriol Parasitol. 2013;4:1.

10. Njunda AL, Njumkeng, Shey C, Nsagha D, Nguedia Assob JC, Kwenti TE. The prevalence of malaria in people living with HIV in Yaounde, Cameroon. BMC Public Health. 2016;16:964.
11. Alemu Abebe, Shiferaw Yitayal, Addis Zelalem, Mathewos Biniam, Birhan Wubet. Effect of malaria on HIV/AIDS transmission and progression. Parasites and Vectors. 2013;6:18.
12. González Raquel, Ataíde Ricardo, Naniche Denise, Menéndez Clara, Mayor Alfredo. HIV and malaria interactions: Where do we stand? Expert Rev. Anti Infect. Ther. 2012; 10(2):153–65.
13. Urban BC, Ing R, Stevenson MM. Early interactions between blood-stage plasmodium parasites and the immune system. Curr Top Microbiol Immunol. 2005;297:25-70.
14. Amexo M, Tolhurst R, Barnish G, Bates I. Malaria misdiagnosis: Effects on the poor and vulnerable. Lancet. 2004;364:1896-8.
15. Cheesbrough M. District Laboratory Practice in Tropical Countries. 2nd ed; 2004.
16. Moody A. Rapid diagnostic tests for malaria parasites. Clin Microbiol Rev. 2002;15(1):66-78.
17. Nkrumah B, Acquah SEK, Ibrahim L, May J, Brattig N, Tannich E, et al. Comparative evaluation of two rapid field tests for malaria diagnosis: Partec Rapid Malaria Test® and Binax Now® Malaria Rapid Diagnostic Test. BMC Infect Dis. 2011;11: 143150.
18. Xiaodong S, Tambo E, Chun W, Zhibin C, Yan D, Jian W, Jiazhi W, Xiaonong Z. Diagnostic performance of CareStart™ malaria HRP2/pLDH (Pf/pan) combo test versus standard microscopy on falciparum and vivax malaria between China-Myanmar endemic borders. Malar J. 2013;12:6.
19. Leke RFG, Djokam RR, Mbu R, Leke RJ, Fogako J, Megnekou R, Metenou S, Sama G, Zhou Y. Detection of the Plasmodium falciparum antigen histidinerich protein 2 in blood of pregnant women: implications for diagnosing placental malaria. J Clin Microbiol. 1999;37:2992–996.
20. Sayang C, Soula G, Tahar R, Basco LK, Gazin P, Moyou-Somo R, Delmont J. Use of a histidine-rich protein 2-based rapid diagnostic test for malaria by health personnel during routine consultation of febrile outpatients in a peripheral health facility in Yaoundé, Cameroon. Am J Trop Med Hyg. 2009;81(2):343–7.
21. Ali IM. Bigoga JD, Forsah DA, Cho‐Ngwa F, Tchinda V, Ama Moor V, Fogako J, Nyongalema P, Nkoa T, Same‐Ekobo A, Mbede J, Fondjo E, Mbacham WF, Leke RGF. Field evaluation of the 22 rapid diagnostic tests for community management of malaria with artemisinin combination therapy in Cameroon. Malar J. 2016;15:31.
22. Tahar R, Sayang C, Ngane Foumane V, Soula, Moyou-Somo R, Delmont J, Basco LK. Field evaluation of rapid diagnostic tests for malaria in Yaounde, Cameroon. Acta Trop. 2013;15(2013): 214-9.
23. Antonio-Nkondjio C, Defo-Talom B, Tagne-Fotso R, Tene-Fossog B, Ndo C, Kengne P, et al. High mosquito burden and malaria transmission in a district of the city of Douala, Cameroon. BMC Infect Dis. 2012;12:275.
24. Nkuo-Akenji T, Ttevoufouet EE, Nzang F, Ngufor N, Fon E. High prevalence of HIV and malaria co-infection in urban Douala, Cameroon. Afr J AIDS Res. 2008;7(2): 229-35.
25. Guidelines for CyFlow counter. Accessed 15 August 2015.
Available: www.sysmex-partec.com
26. Koanga Mogtomo ML, Kojom Foko LP, Assokom Okoubalimba EV, Embolo EE, Ngono Ngane AR. High risk of transfusion-transmitted malaria (TTM) from student blood donors living in the town of Douala, Cameroon. J Clin Infect Dis Prac. 2016; 1:108.
27. ACCUCARE one step Malaria Pf/Pv Ab detection Test guidelines. Accessed 30 September 2016.
Available: www.labcarediagnostics.com
28. Cohen J. A coefficient of agreement for nominal scales. Educ. Psychol. Meas. 1960;20:27-46.
29. Amuta, EU, Houmsou, RS, Diya AW. Malarial infection among HIV patients on antiretroviral therapy (ART) and not on ART: A case study of Federal Medical Centre Makurdi, Benue State, Nigeria. Asian Pac J Trop Biomed. 2012;S378-S381.
30. Omoti CE, Ojide CK, Lofo PV, Eze EE, Joy C. Prevalence of parasitemia and associated immunodeficiency among HIV-malaria co-infected adult patients with

highly active antiretroviral therapy. Asian Pac J Trop Med. 2013;126-30.

31. Iroezindu MO, Agaba EI, Okeke EN, Daniyam CA, Obaseki DO, Isa SE, et al. Prevalence of Malaria Parasitaemia in Adult HIV-infected Patients in Jos, North-central Nigeria. Nigerian J Med. 2012;21(2):209-13.
32. Tay Sammy CK, Badu K, Mensah AA, Gbedema SY. The prevalence of malaria among HIV seropositive individuals and the impact of the co- infection on their hemoglobin levels. Ann Clin Microbiol Antimicr. 2015;14:10.
33. Van Geertruyden J-P, Mulenga M, Kasongo W, Polman K, Colebunders R, Kestens L, D'Alessandro U. CD4 T-Cell count and HIV-1 infection in adults with uncomplicated malaria. J Acquir Immune Defic Syndr. 2006;43(3):363-7.
34. Chavale H, Santos-Oliveira JR, Da-Cruz AM, Enosse S. Enhanced T cell activation in *Plasmodium falciparum* malaria-infected human immunodeficiency virus-1 patients from Mozambique. Mem Inst Oswaldo Cruz, Rio de Janeiro. 2012;107(8):985-92.
35. Suchita JL, Krunal KS, Pragnesh PP. Comparative study of rapid malaria antigen test with Giemsa stained peripheral smear for diagnosis of malaria. Med Sci. 2013; 2(6):509-510.
36. Khatib Y, Gulhane S, Sequeira K. Correlation and comparative evaluation of thick smear, thin smear and antigen detection test in the diagnosis of malaria. Sch J App Med Sci. 2016;4(5E):1770-3.
37. Ayele DG, Zewotir TT, Mwambi HG. Semiparametric models for malaria rapid diagnosis test result. BMC Public Health. 2014;14:31.
38. Ayele DG, Zewotir TT, Mwambi HG. The risk factor indicators of malaria in Ethiopia. Int J Med and Med Sci. 2013;5(7):335-47.
39. World Health Organization. Malaria rapid diagnostic test performance. Summary of results of WHO products testing of malaria RDTs: round 1-5 (2008-2013). Available:www.who.int (Accessed 23 September 2015)
40. Rogier C, Henry M-C, Trape J-F. Evaluation épidémiologique du paludisme en zone d'endémie. Med Trop. 2009;69 : 123-42.

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/17634