

Full Length Research Paper

Malaria among relatives escorting sick patients during the dry season to Karume Health Centre, Mwanza, Northwestern Tanzania

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Malaria is still a public health problem in the world. It accounted for an estimated 214 million cases and 438,000 deaths in the year 2015. During the dry season, most people are likely to be asymptomatic and therefore fail to be diagnosed with malaria. This study established the proportion of people who came to health facility, escorting sick relatives and had detectable malaria parasites. This was a cross sectional study. All relatives who escorted sick patients to Karume Health Centre between August and December 2013 were screened for malaria using malaria rapid diagnostic test (mRDT) and single round PCR targeting mitochondrial DNA. A total of 400 relatives were screened for malaria using two methods. Prevalence of malaria was 14.5 and 16.8% by mRDT and polymerase chain reaction (PCR), respectively. The prevalence of malaria was higher among febrile patients by methods, mRDT (17.8%) and PCR (17.1%), respectively. The prevalence of asymptomatic malaria was 16.4 and 16.5% by mRDT and PCR, respectively. The overall agreement between the two tests was 87.1% with positive agreement of 63.8% and negative agreement of 91.2%. There were a substantial proportion of patients with malaria who visited the health facilities during the dry season. mRDT and single round PCR targeting mitochondrial DNA had a good agreement and can be used for detection of both symptomatic and asymptomatic malaria. Provider initiated screening can help to improve malaria detection during the dry season, as we move towards reduced malaria prevalence and elimination phase.

Key words: Asymptomatic malaria, prevalence of malaria among relatives, mitochondrial DNA, polymerase chain reaction (PCR).

INTRODUCTION

Malaria is still a public health problem especially in the WHO African region. Form 2015, the African region

accounted for 88% of the 214 million cases that occurred in the world (WHO report, 2015). The trend however

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shows that malaria is declining globally from 262 million cases in 2000 to 214 million cases in 2015 (WHO report, 2015). The decrease is attributed to the use of Insecticide treated bed nets (ITNs), indoor residual spraying and efficacy of artemisinin based combination therapies (ACTs).

In most regions including Tanzania, malaria transmission occurs mainly during the rainy season and is lowest during the dry season (Oesterholt et al., 2006). During the dry season, there are usually few cases of malaria and morbidity and mortality goes down to close absence of the disease depending on endemicity of malaria (Oesterholt et al., 2006; Ardiet et al., 2014). During the dry season therefore, most individuals are likely to be asymptomatic while carrying the parasites.

Asymptomatic patients far outnumber the symptomatic infections (Lindblade et al., 2013; Alves et al., 2002; Lin et al, 2014) and are likely not to come to hospital. These infections may last from weeks to months and to an average of 194 days (Males et al., 2008; Bottius et al., 1996; Felger et al., 2012). It has been reported that proportions of individuals remain with the parasites in blood circulation and can be transmitted at the beginning of the next rainy season.

Studies in Africa have clearly demonstrated that, asymptomatic cases harbour gametocytes and can therefore be a significant transmission reservoir (Dunyo et al., 2006; Bousema et al., 2004). Another challenge during the dry season is the diagnostic method used for screening (Ardiet et al., 2014; Permeger et al., 2006). mRDT is now widely accepted in malaria endemic countries but its sensitivity when parasitaemia is low is a limiting factor. Existence of submicroscopic parasites (Kamugisha et al., 2012; Lindblade et al., 2013) makes the gold standard test microscopy to be of limited value, during dry season when parasites loads are low and patients are asymptomatic. Thus investigating the utility of other sensitive methods such as single round PCR targeting, mitochondrial DNA is important in the era of declining malaria when elimination is becoming an agenda.

In this study we screened people who come to hospital, not seeking treatment for themselves but escorting relatives who are sick. The aim was to see what proportion of these relatives actually reaches a service providing facility and has fever as, malaria infection. This is a form of active case detection (Van Eijk et al, 2016) that is utilizing relatives and siblings at the health facility.

MATERIALS AND METHODS

Study area and design

This was a cross sectional study conducted at Karume Health centre in Mwanza city, Northwestern part of Tanzania. The area is mesoendemic lying in Lake Victoria basin at an altitude of 1140 m above sea level. The basin is endemic for malaria with perennial transmission. The catchment area of Karume health centre is about

40,000 inhabitants.

Study participants and recruitments

This study recruited all relatives and fellow siblings who escorted their relatives to receive care at Karume health centre. All relatives who escorted their sick families to the centre during the study period were informed about the study and those who agreed signed an informed consent. Those who consented were interviewed using a structured questionnaire, intending to collect the social demographic characteristics and the use of antimalarial drugs in the past two weeks prior to recruitment. A clinical examination was done to the participants and blood taken for mRDT were dried in blood spot for PCR investigation.

Parasitological examination

The presence of malaria parasites was determined using mRDT and PCR targeting parasite mitochondrial DNA. mRDT was performed immediately at Karume health centre using SD Bioline Malaria Ag Pf/Pan mRDT, Korea. The test was done according to manufacturer's instructions. PCR targeting parasite mitochondrial genes was done in Uppsala University in Sweden. The method used was as described previously (Haanshuus et al., 2013). Only a single round PCR was done contrary to the nested PCR usually performed for DNA analysis of other malaria parasite genes. It has been stated previously that, due to high number of mitochondria per parasite, a single round PCR is enough to detect even small amounts of DNA.

Data analysis

Data entry, cleaning and analysis was done using SPSS version 17 software. Frequencies and cross tabulation were done to obtain proportions and chi square for associations of the study variables. Due to comparison of two tests without a gold standard, Cohen's kappa test was used. P-value of less than 0.05 at 95% confidence interval was considered significant.

Ethical clearance

The study was approved by joint CUHAS/Bugando ethics committee. Permission to conduct the study at Karume health centre was sought from other relevant administrative authorities. Participants above 18 years signed an informed consent before participating in this study. For those below 18 years, an informed assent was sought together with informed consent from the guardians/parents. All patients who were diagnosed with malaria were treated with artemether-lumefantrine (ALU) accordingly. Those who had no malaria but were febrile were investigated for the cause of fever and treated.

RESULTS

Social demographic characteristics

A total of 400 relatives of patients who attended Karume health centre were recruited. Females were the majority with 333(83.3%). The youngest participant was 2 years old while the oldest was 90 years with mean age of 30.2. The majority 367(91.8%) of participants were adults

Table 1. Socio-demographic characteristics of the study participants.

Variable	Number	Percentage
Age group		
≤5 years	4	1.0
6-10 years	5	1.3
11-18 years	24	6.0
<18 years	367	91
Sex		
Female	333	83.3
Male	67	16.8
Fever		
Yes	152	38.0
No	248	62.0

Table 2. Comparison of mRDT and single round PCR targeting mitochondria DNA.

Parameter	mRDT positive	mRDT negative	Total
PCR positive	37	30	67
PCR negative	21	312	333
Total	58	342	400

above 18 years (Table 1).

Prevalence of malaria

Among the 400 participants, 152(38%) had fever (body temperature >37.5°C) on the day of recruitment. The overall prevalence of malaria by using mRDT was 58(14.5%). The prevalence was higher among those who were febrile at recruitment 23(17.8%), compared to those who were afebrile (asymptomatic host malaria) 35(16.4%), though the difference was not statistically significant (p-value 0.79) (Table 2). The prevalence by, single round PCR targeting mitochondrial DNA was 67(16.8%), asymptomatic relatives was 16.5% and febrile relatives was >17.1%. The prevalence was higher among febrile relatives than those who were afebrile and the difference was statistically significant (p-value 0.022).

Comparison of mRDT and mitochondrial DNA tests

When the two diagnostic tests were compared (Table 2) in the absence of a gold standard test, the overall percent agreement between mRDT and PCR was 87.1% and p-value was <0.001. The positive agreement was 63.8% and the negative agreement was 91.2%. There was a

good agreement between the two tests by Cohen's kappa of 0.5.

Prevalence of fever with age

When the participants were divided into different age groups, the prevalence of fever declined with age. The prevalence was 100% in the age group <5 years and lowest in the adult group of 18-90 years. The difference was statistically significant (p-value = 0.004).

DISCUSSION

Prevalence of malaria by mRDT

The overall prevalence of malaria was high considering that, this study was done during the dry season when transmission is low. There have been a different prevalence's of malaria during the dry season but it is known that, a significant proportion of individuals remain with microscopically undetectable parasites in blood which form a reservoir for the next transmission.

The prevalence of asymptomatic malaria among patients was 16.4%, which was higher than that recently reported in school children in Tanzania (Nzobo et al., 2015). This is almost similar to the previous study in school children in a nearby place, which was 14.3% (Mazigo et al., 2010). In different parts of Africa, the prevalence of asymptomatic malaria varies considerably and prevalence up to 80% was reported in Cameroon (Okell et al., 2012).

Comparison of mRDT and mitochondrial DNA

A gold standard method for diagnosing malaria is microscopy. During the dry season and especially in asymptomatic patients, microscopy may be a limited diagnostic test. Existence of sub-microscopic parasites in patients treated effectively with ALU in this area has been shown (Kamugisha et al., 2012), while those in other areas has also been reported. The use of mRDT and PCR during the dry season in this study gave a prevalence that is similar to other places. There was also a good agreement between the two diagnostic tests carried out, and the result is comparable to the findings in other studies.

The single round PCR is also being shown to be effective, in the field for screening of malaria parasites during the dry season. Due to shorter time used and its good agreement with nested PCR in this study and its specificity and sensitivity documented previously (Haanshuus et al., 2013), it is clear that mRDT can be used for screening and diagnostic purposes in endemic countries. Deploying different diagnostic methods for asymptomatic, malaria infection is vital and has been

suggested previously (Bousema et al., 2014).

Prevalence of malaria by mRDT with age

This study shows that, by doing active case detection among relatives escorting patients with malaria, it is likely to increase detection of malaria cases. All children aged <5 years, who visited the clinic as siblings to other sick children were also febrile but their fever was not reported. In the higher age group as well, the relatives/siblings also had fever, which was not reported. The patients who were positive for malaria and other diseases such as urinary tract infection and upper respiratory tract infection were treated. Those who received an antimalarial dose might have a reduced risk of coming at a later date, with severe malaria and/or uncomplicated malaria probably associated with other complications such as anaemia.

The main reason for parents coming and complaining or intending to treat only one child could be due to limited resources enough to treat all family members with fever. The limited funding for cost sharing and purchasing drugs are then concentrated to one person, who looks to be more severely ill than others. This study did not look at the factors that were making febrile individuals not to report their illness but recommend such a study in this area. The hidden point may be that, due to limited funding, the treatment given to the sick individual may be divided and utilized by the other sick relatives at home. If this occurs then, failure to complete the required ALU dose and other drugs are likely to occur in both relatives and this may lead to quick building up of drug resistance.

Data from this study shows that mimicking the strategy used by the same health facilities for HIV testing that is known as Provider initiated HIV counselling and testing (PICT), might also work for malaria. Although in PICT for HIV, the testing is for patients coming with other diseases but in lower facilities testing for relatives especially during low transmission, season might be another way of doing Focused Screening and Treatment (FSAT) and hence help in the control against malaria.

Conclusion

The prevalence of malaria among relatives visiting the health centre was low, but there were a high percentage of relatives with fever who does not complain about it at health facility. The prevalence among asymptomatic relatives was also low. The children accompanying their family members are also having complaints and malaria. There was good detection of malaria by both mRDT and PCR targeting mitochondrial DNA.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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