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MALARIA MICROSCOPY IN PRIMARY HEALTH CARE CENTERS IN KHARTOUM STATE, SUDAN: EXTERNAL QUALITY ASSURANCE STUDY
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S. A. Mustafa and A. S. Aljafari

ABSTRACT

Background: **Adequate diagnosis of Malaria is achieved by detection and identification of malaria parasites through examination of Giemsa's stained blood films by competent personnel.**

Objective: **To identify the competency of laboratory personnel of the clinical laboratories attached to primary health care centres at Khartoum State- Sudan, by applying External quality assurance (EQA) slide check.**

Design: **A cross-section and facility based study conducted during the period from October to December 2015.**

Setting: **Primary health care centres at Khartoum State, Sudan.**

Subjects: **The study enrolled 52 clinical laboratories.**

Result: **For the EQA slides, there were 262 positive slides, 199 of them were P. falciparum and 63 were P. vivax. Study subjects achieved 213 true positive results and 204 true negative results, sensitivity and specificity were 81.3%, 79% respectively. For species detection, they reported 134 true positives and 54 true negatives for P. falciparum (sensitivity 84.8%, specificity 98%); and 55 true positive and 134 true negative P. vivax (sensitivity 98%, specificity 84%). False negative results tend to be obtained when parasite load in scanty (OR: 3.64), and is likely associated with P. falciparum (OR: 1.9). Conclusion: This study concluded that the quality of malaria microscopy for the study facilities was poor and the personnel were found of low competency. Poor infrastructure, heavy workload, and absence of training were among the factors that affect the competency of study personnel.**

INTRODUCTION

Malaria is a febrile disease caused by protozoan blood parasites species belong to the genus Plasmodium and it is transmitted through an infective mosquito bite. Malaria affects 303 million of the 2.5 billion individuals at risk worldwide. It kills more

than one million annually, 90% of them are in Africa and almost half of them are children (1). Two Plasmodium species are prevalent in Sudan, P. falciparum (90%) and P. vivax. In Sudan, Malaria is responsible for annual 7.5 cases, 35000 deaths, and 11% of hospital visits. During the rainy season in high transmission areas, the frequency of malaria reaches 85% (2- 4). Beside the 2,877,000 disability-adjusted life years (DALY) (5), malaria costs the local and international community a fortune of US$81 million which was dispelled out to roll-back malaria in Sudan in 2015 (6).

Proper case management depends mainly on prompt and accurate diagnosis. Early diagnosis and treatment are important because untreated malaria may be associated with serious life-threatening complications (7).

Guidelines issued by WHO for malaria management in endemic countries recommended that treatment should be based on laboratory confirmation through microscopic detection and identification of the parasite (8). This is achieved be strictly quality controlled parasite-based microscopic detection and identification of the parasites (Malaria microscopy). Rapid diagnostic tests (RDTs) are also of a greater value especially for screening, however, they are only additional tools (9,10). Excessive reliance on RDTs may cause overlooking potentially severe non-malarial febrile illnesses (11,12). Polymerase chain reaction (PCR) could be effective only in low transmission areas (13).

Malaria diagnostic tests in endemic areas should meet several standard measures. They should be rapid; reliable; feasible; sensitive and specific; and cost-effective. Decades of researches revealed that malaria diagnostic methods usually accompanied by several limitations.

Microscopic examination of Giemsa's stained blood films (thin and thick) is the gold standard method for detection and identification of malaria parasite. They need to be adhered to strict quality measures to ensure their effectiveness (10,14). Inadequate or failure of parasitological confirmation may lead to serious clinical consequences (15). Poor adherence of the doctors to the laboratory confirmation of malaria may also lead to the same effect (16,17).

Nothing but chemotherapy is applied for case management. Vector control is the famous approach for control, as there is no effective preventive measure so far; Malaria vaccine is still far away (14). In this contribution, we tried to verify the role of laboratory personnel in the dilemma of malaria diagnosis in Sudan. We implemented an external quality assurance (EQA) program for randomly selected and representative clinical laboratories at Khartoum State to assess the competency of laboratory personnel. The program included cross-checking of known malaria slides (EQA).

MATERIALS AND METHODS

This is a cross-sectional and facility-based study of a qualitative approach conducted during the period from October to December 2015. The objective of this study was to identify the competency of laboratory personnel for malaria microscopy by using on-site evaluation by External Quality Assessment (EQA) slides. Fifty-two clinical laboratories in Khartoum State were enrolled in this study. Laboratories were randomly selected to represent the 7 Municipals of the State. External quality assessments (EQA) slide panels were prepared. Every panel box contains 10 slides with known positive and negative Giemsa's stained blood films. Positive slides contain species (P. falciparum and P. vivax) with different stages and different parasite densities. Every laboratory was given an EQA slide box for microscopic examination. Results were collected and then compared

with the actual EQA slides. 520 records were obtained from the subjects. Data tabulation and management were made using IBM® Statistical Package for Social Sciences (SPSS v 22). Sensitivity, specificity, and negative and positive predictive values were identified.

Ethical consideration:

This study received ethical approval from the Ethical Committee of the Faculty Board Council (Faculty of Medical Laboratory Sciences- Al Neelain University, Khartoum). Permission to conduct the study was granted by the General Administration of Health insurance, the responsible body for the primary health care centres. Informed consent was obtained from each participant.

RESULTS

This study enrolled 52 clinical laboratories from the semi-governmental health care centres at Khartoum State. For the EQA slides, there were 262 positive slides, 199 of them were P. falciparum and 63 were P. vivax. Study subject achieved 213 true positive and 204 true negative results. The sensitivity and specificity were: 81.3%, 79% respectively, and positive predictive value was 3.94% (table 1). For species detection, study subjects reported 134 true positives and 54 true negatives for P. falciparum (sensitivity 84.8%, specificity 98%); and 55 true positive and 134 true negative P. vivax (sensitivity 98%, specificity 84%) (Table 2). False negative results tend to be obtained when parasite load in scanty (+1), OR 3.64 (table 3), and likely associated with P. falciparum (OR, 1.9) (table 4).

Table-1

Comparison between Results obtained by participants (subject results) and results of the EQA slides

		EQA slides positive	Negative	Total
Subject results	Positive	213	54	267
	Negative	49	204	253
Total		262	258	520

Table-2

Comparison between parasite species of EQA slide and results obtained by study subjects

		EQA slides P. falciparum	P. vivax	Total
Subjects results	P. Falciparum P.	134	1	135
	vivax	23	55	78
Total		157	56	213

Table-3
Comparison between parasite density of EQA slide and results obtained by study subjects

True Positive		Subjects results False negative		Total
Parasite density (EQA slides)	+1	2	45*	47
	+2	147	4	151
	+3	28	0	28
	+4	36	0	36
Total		213	49	262

** OR (3.64)*

Table- 4
Comparison between parasite species of EQA slide and results obtained by study subjects

True Positive		Subjects results False negative		Total
Species (EQA slide)	*P. Falciparum*	157	42* 7	199
	P. vivax	56		63
Total		213	49	262

** OR (1.9)*

DISCUSSION

Detection and identification of malaria parasites with estimation of parasite density is the proper method for diagnosis and leads to effective case management. This is usually achieved by examination of Giemsa's stained thick and thin blood films, which is most specific, sensitive, and cost-effective method. However, malaria microscopy is usually accompanied by several limitations. Failure of adherence to standard quality measures for preparing and staining of the blood films may lead to misdiagnosis. There are many other factors that negatively affect the quality of malaria microscopy, such as poor education and training of the laboratory personnel. In this study we assessed and evaluated the competency of the laboratory personnel working at 52 laboratories attached to the Primary Healthcare centres in Khartoum State using on-site EQA slide at the actual condition

For the detection of malaria parasite, the positivity rate obtained by the study personnel was 80%, and this is less than the minimum competency level suggested by the WHO which is 90% (8). However, the participants achieved the minimum requirement for the specificity (obtaining true negative results) which is 80%. This finding indicates that the study participants have the tendency to issue false positive results.

For species identification, the participants reported a sensitivity of 84% for identifying P. falciparum, which is less than the minimum of the competency level (90%). They reach the higher level of specificity (98%) in comparison with the minimum competency level designated by the WHO (90%) (8). These measures were typically

opposite for P. vivax. The hallmark of these findings is that the participants detect P. vivax in an acceptable competency but they failed to identify them correctly; and they couldn't detect P. falciparum in an acceptable competency, while they achieved the maximum competency for the identification. We believe that, the workload, lab environment, and the treating doctors, all put a pressure on the laboratory personnel pushing him towards issuing positive results. Also, it should be put in consideration that P. falciparum constitutes more than 90% of the infections.

The results also revealed that the participants tend to obtain false negative results when the parasite density is scanty. Scanty parasitemia usually indicates that the parasite density is less than 10 parasite cells per 100 fields (x100 objective); which is equivalent to parasite count less than 100 parasite cell/μl. The WHO advises that the individual must examine 150200 good fields (x100 objective) of Giesam's stained thick blood film before issuing negative result. The competent individual, when given the suitable time, can detect malaria parasite even when the parasite count is low as 20 parasite cell/μl.

It is evident from the results that false negative results are associated with P. falciparum in low density. This should not be attributed only to the low competency of study personnel. Small size and delicate appearance of P. falciparum ring stage (which the predominant stage in the peripheral blood) render this stage difficult to detect especially when found at scanty numbers.

This study enrolled clinical laboratories of the primary healthcare centres, which are semi-governmental facilities. They offer services that are covered by governmental health insurance. The majority of the visitors of these facilities are from the middle and lower class; they are poor and can't afford better health care services provided by the private sector. These primary health care centres are usually crowded and deficient of many of the essential infrastructures. The laboratory environment is not good enough to relax the personnel for doing the job perfectly. The workload is too heavy and the human resources are scarce. Poor working environment and heavy workload are negatively affect the capacity and competency of the laboratory personnel for of malaria microscopy (18, 19).

The EQA slide programs were found to improve the efficacy of malaria microscopy to obtain high sensitive and specific results. For the EQA to be effective, it should be followed by a reform program, and should be associated with an incentive action to reinforce good practice. The infrastructure of the clinical laboratory should provide the minimum requirements of safety and relaxing environment, together with the suitable workload and workflow.

There should be a mandatory in-service training program for the lab personnel. It is recommended by many studies that, internal and external quality control programs in malaria microscopy should be mandatory for any health care facility assigned for malaria diagnosis (20-22). Furthermore, there is a loud sound trend for licensing the practice of malaria microscopy (23).

We are not defending the study personnel; we do agree that they are of an unacceptable quality. However, they are working in an environment where the quality of malaria microscopy could not be improved.

Why couldn't malaria microscopy be as importance as reading cytopathology or histopathology slides?

CONCLUSION

This study concluded that the EQA check revealed that the quality of malaria microscopy for the study facilities was poor and the personnel were of low competency. Poor infrastructure, heavy workload, and

absence of training were among the factors that affect the competency of study personnel.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTION

Sayed A. Mustafa:

- Conception and design
- Acquisition of data
- Interpretation of data • Discussion

Alfatih S. Aljafari

- Conception and design
- Analysis and interpretation of data;
- Discussion
- Drafting the manuscript

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