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COMPARISON OF RELIABILITY OF RAPID DIAGONOSTIC TEST TO MICROSCOPY IN THE DIAGNOSIS OF MALARIA AT GULU REGIONAL REFERRAL HOSPITAL

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ABSTRACT

***Background*: Malaria is one of the leading causes of mortality in sub-Saharan Africa and continues to be a threat to life. Everyday 320 people die due to malaria in Uganda. Statistics from the ministry of health show that of all outpatient visits at health Centres, malaria represents 25 - 40 % with 9 - 14 % in in-patient. Microscopy remains the main stay for malaria diagnosis. Nevertheless it is slow and needs a lot of experience and expertise, consequently there is need to use a faster method but retaining the precision of the microscope.**

***Objective*: To determine the reliability of the rapid diagnostic test in comparison to microscopy in the diagnosis of malaria thereafter improve the management of malaria.**

***Design*: An experimental study.**

***Setting*: Gulu Regional Referral Hospital laboratories.**

***Subjects*: The study was experimental; using 250 malarial suspected participants with fever .The malaria rapid diagnostic method and microscopy of the stained malarial slides were the methods used.**

***Results*: Of the 250 samples, 214 were found to be negative using microscopy and 208 using Rapid Diagnostic Test suggesting over suspicion of malaria. Microscopy had a sensitivity of 85.7%, specificity of 94.7% and Negative predictive value (NPV) of 97.2%.**

***Conclusion*: RDTs can be used to quickly confirm the clinical diagnosis of malaria to reduce irrational use of anti-malarials when microscopy is not available before initiating treatment to avoid irrational use of drugs. However due to the cost, microscopy still remains the gold standard method for the diagnosis of Malaria.**

INTRODUCTION

Today, malaria is responsible for more illness and death than any other single disease in Uganda (1,2,3). Malaria is highly endemic with 63% of the population exposed to high transmission levels and 25% exposed to moderate transmission levels while 12% live in areas of low transmission or unstable malaria transmission that are epidemic prone (1,2,3). The 2005-2006 Uganda National Household Survey (UBOS) revealed that half of the population that fell sick reported malaria or fever as their major symptom for illness (4).

In Uganda, malaria is more prevalent during the rainy season of March to June and August to November, with exception of Northern Uganda where the malaria season is more prevalent between May and November (1,4). Early and accurate diagnosis of malaria followed by prompt treatment reduces the risk of severe disease in malaria endemic areas (5, 6). Presumptive treatment of fevers with antimalarial drugs is widely reduced malaria morbidity and mortality especially at lower health facilities where microscopy is not readily available (5,6). Recent advent of rapid diagnostic tests (RDT) for malaria may be a significant step forward in case detection, management and reduction of unnecessary treatment (7) Such RDT could also be useful in malaria diagnosis during population-based surveys and to provide immediate treatment based on the results. However, the accuracy of RDT under field conditions in low transmission areas remains questionable (7).

MATERIALS AND METHODS

This was an experimental study. The area of study was Gulu Regional Referral Hospital laboratories. This hospital is situated in Northern Uganda in Gulu municipality-Laroo division and caters for patients from the surrounding areas and those referred to it from other health facilities. All patients with a preliminary clinical diagnosis of malaria and needed to be tested for malaria parasites during this period wereincluded inthe study.

Using the Keish and Leslie (1995) formula; with 'p' (prevalence of malaria in Uganda) value of 24.2% we obtained a sample size of 250 people (8, 9). Then systematic sampling technique was used, the first person was picked randomly, and then every 3rd person was then included in the sample.

Capillaries finger blood samples were collected aseptically from the participants in the study who were sent for malaria testing and tested for malaria parasites using rapid diagnostic test and thick and thin blood smears were prepared for microscopy. The blood sample was put on a clean slide and air dried. This was then immersed in field stains A, rinsed in water, then immersed again in Field stain B rinsed in water before air drying and observed under the microscope after applying an oil immersion.

Data were entered, purified and analysed using SPSS17.0 and presented in form of tables, graphs. The quality control of data was ensured with the assistance of laboratory technician doing microscopy, the microscope pretested and researchers briefed and trained on the use of RDTs whose quality was ensured by observing proper manufacturer's instructions as regards use and storage.

The proposal was presented to the Faculty IRB and hospital IRB and approved. Informed consent of the participants was sought. Confidentiality was upheld during the whole research process especially in dealing with the samples.

The findings of the study were disseminated to the academic board Gulu University, Head of Department Public Health, Head of the Research Group, Gulu Regional Referral Hospital the Researchers and Gulu University Library.

RESULTS

Table

The number of participants according to their sex

Sex	frequency	Percentage
Female	159	63.6
Male	91	36.4
Total	250	100

It can be observed that out of the 250 participants who participated in the study 159(63%) where female and 91(36.4%) were male. The mean age was 23.2 years (range 2weeks to 81 years). This correlates to what is widely known that females have a good health seeking behaviour compared with their male counterparts. At least 62(24.8%) had taken some medications before reporting which included anti-malarials, anti-biotics, analgesics and local herbs

Table 2

Sshows the results obtained from a paired sample t test of Microscopy and RDT.

	N	Correlation	Sig.
Pair 1 Microscopy results & RDT results	250	.695	.000

Paired Samples Correlations: p<.001, Confidence interval of 95%The above table of correlation shows a strong positive correlation between Microscopy and RDT tests with a significant p-value of p<.001 and a confidence interval of 95%.

Table 3
Results of microscopy

	Results		
Diagnostic tests	Positive	Negative	Total
Microscopy	36(14.4%)	214(85.6%)	250

Table 4
Results of RDT

	Results		
Diagnostic test	Positive	Negative	Total
RDT	42(16.8%)	208(83.2%)	250

Table 5
Cross tabulation of microscopy results with RDT

			RDT results		
			Negative	Positive	Total
Microscopy results	No malaria parasites seen	Count % within Microscopy results	202 94.4%	12 5.6%	214 100.0%
	(+) Malaria parasites seen	Count % within Microscopy results	6 37.5%	10 62.5%	16 100.0%
	(++) Malaria parasites seen	Count % within Microscopy results	0 .0%	11 100.0%	11 100.0%
	(+++)Malaria parasites seen	Count % within Microscopy results	0 .0%	9 100.0%	9 100.0%
Total		Count % within Microscopy results	208 83.2%	42 16.8%	250 100.0%

It can be observed that 12 (5.6%) results of the 214 cases reported negative by microscopy were found to be actually positive by the RDT. This is because RDTs detect antigens which might persist in the body after the parasites observed by microscopy have been cleared. HRP2 actually persist in circulation for up to one month post serum parasite clearance. This occurs in patients who have been taking antimalarials and the parasites levels are low hence undetectable on microscopy

Of the 16 results reported positive (+) for Malaria parasites using microscopy, 6 (37.5%) were negative using RDT. This is because of low parasite density hence a negative RDT result.

We found out that all cases of (++) and (+++) with microscopy were also positive using RDT. This shows that at high parasite densities, the sensitivity of RDT is very high and compares to that of microscopy.

Table 6
Sensitivity and specificity of RDT using microscopy as the gold standard

	Positive	Negative
True	30	202
False	12	6

Sensitivity:
True positive
[True positive +false negative]
= 30/(30+6)
= 83.3%
Specificity:

True negative
[True negatives + false positives]
=202/214
=94.4%
Negative predictive value (NPV)
True negative
[True negative + false negative]
= 202/(202+6)
=97.1%

DISCUSSION

The accuracy of clinical diagnosis of uncomplicated malaria was compared using microscopy and RDT (Paracheck). The diagnostic accuracy was compared with microscopy as the gold standard with sensitivity (85.7%), specificity (94.7%) and a NPV (97.2%) of Paracheck in this study. Compared with Batwala *et al*., 2010 whose research showed a sensitivity of 91.0% and specificity of 86.7% and NPV of 95.8%, a

number of factors might have contributed to the low sensitivity of RDT including existence of low density infections which was evident with a parasites density of plus one (+) which showed 6(37.5%) negative test results by paracheck and yet confirmed to be positive by microscopy (5, 6).

However, in another study the RDT sensitivity and specificity were found to be low with values of 88.6% and 88.2% respectively. This conforms to our findings which also demonstrated a low sensitivity (83.3%). In that study it was found that RDTs reduced antimalarials dispensing from 98.9% to 32.1% in cases aged five years and above hence reducing on irrational drug use significantly (10,11).

Microscopic detection of malaria parasites in thick blood smears remains the most appropriate diagnostic method in endemic countries however this is difficult as microscopy is labour intensive and needs regular supply of consumables like stains. These factors affect the wide spread use of microscopes (5, 6).

Uganda has adopted RDT as a method for parasitological diagnosis of malaria in addition to microscopy (1, 2, 3). RDTs are rolled out in lower level HCs (Health centers) where microscopy services are not functional or not available. If a study of RDTs is guaranteed, the distribution should be extended to lower level facilities. In addition, it is vital to routinely evaluate the performance of RDTs as they are being rolled out in the country (1, 2, 3). RDTs could enable the reduction in the cost of antimalarials as fewer drugs will be needed because less people will be confirmed with malaria (12, 13). RDTs do not require a lot of skilled labor compared to microscopy and there is minimal intra observer discrepancy. It was observed in this research study that the results obtained by using RDTs were very reliable and could be confidently used to quickly initiate treatment in order to avert the mortality associated with malaria. It can also be inferred from this study that there is a significant overlap in the signs and symptoms of malaria and other diseases which health care providers should be aware of and where possible should always correspond their suspicion with the results of a diagnostic test like RDT(14).

CONCLUSION

This study showed that RDT has a sensitivity at high parasite densities (++ and +++) and low sensitivity at low parasitic density (+). Therefore, it is as reliable as microscopy in the diagnosis of malaria at high parasite densities. High specificity of the RDT in this research means that most of the patients without malaria were correctly detected. Thus making use of RDTs in the diagnosis of malaria can significantly reduce irrational drug use and proper management of patients. Where affordable we recommend that HRP2-baesd RDTs can be used in centers without microscopes or personnel to quickly confirm the clinical diagnosis of malaria before initiating treatment which will help to reduce the irrational use of antimalarials and subsequent development of resistance.

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