Original Research Article

Comparison of rapid test and conventional staining methods for the diagnosis of malaria in a tertiary care hospital

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Abstract

Rapid diagnosis is extremely important for effective treatment and reducing the mortality and morbidity of malaria. Therefore, study was undertaken to compare the efficacy of conventional staining methods (Leishman stain, Field's stain) with rapid diagnostic tests (Advantage Malaria Card Test)for diagnosis of malaria keeping leishman stain as a gold standard test. The present study was conducted from January 2016 to December 2016. Of 524 cases studied, 37 cases were positive for malarial parasites by Leishman stained smear and Advantage Malaria Card Tests.13 cases were infected with P.falciparum and 24 with P.vivax infection. Advantage Malaria Card Test showed 100% sensitivity and specificity each ascompared to gold standard test. Rapid diagnostic tests showed good performance as a screening test. Therefore it can be recommended for wide-scale usage when microscopy is not available and immediate clinical diagnosis is required.
Key Words: Malarial parasite, Peripheral smear, Rapid Diagnostic Test.

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INTRODUCTION

Malaria is an age-old disease and is associated with human kind since time and contributes the most to the global mortality and morbidity[1]. Resurgence has been in many parts of the globe due to drug resistance and non-availability of anti-malarial vaccine[2]. There arevarious methods currently available for identification like light and fluorescence microscopy, immuno-chromatographic assays, serology and nucleic acid amplification techniques (NATs)[3]. Other known techniques are loop mediated isothermal amplification (LAMP)[4], flow cytometry[5] etc. However, the most commonly tests are limited still to microscopy or rapid diagnostic tests. Conventional methods (Leishman stain or Giemsa stain)still remained the gold standard for the diagnosis of malaria. However these tests requires expertise, are labor-intensive and time consuming.[6] Various rapid techniques like PCR, DNA probe assay are available for malaria diagnosis but are impractical for routine laboratory use.[7,8]. Therefore an attempt was made to compare the efficacy of conventional staining methods (Leishman stain, Field's stain) with rapid diagnostic tests.

AIM AND OBJECTIVES

Comparison of rapid test with that of conventional peripheral smear in diagnosis of suspected cases of malaria.

MATERIAL AND METHODS

The present study was carried out in the Microbiology Department, Chirayu Medical College and Hospital, Bhopal for a period of one year (January 2016 to

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December 2016). The study was approved by Institutional ethics committee.

Inclusion Criteria: Clinically suspected cases of both sexes and all ages whose samples (5ml in EDTA) were received in the microbiology laboratory.

Exclusion Criteria: Patients already on anti-malarial drugs were excluded from the study. Thick and thin smears were prepared on slides one each for Leishman and Fields stain.[9] All the samples were then subjected to antigen detection using Advantage Malaria card test (Pf and Pv) kit. Advantage malaria is an immunoassay based on the "sandwich" principle for rapid qualitative determination of malarial antigen (pLDH and/or HRP-2) in human blood as an aid in the diagnosis of malaria. Tests were performed as per instructions provided in the kit. All the results of conventional staining methods and rapid antigen detection test were evaluated and compared for the diagnosis of malaria.[10]

OBSERVATIONS AND RESULTS

Table 1: Percentage positivity of Malaria Cases (n=37)

Species	Number Positive	Percentage
Plasmodium Falciparum	13	35.13%
Plasmodium Vivax	24	64.86%
Total	**37**	**100%**

Table 2: Age wise distribution of positive malaria cases

Age in years	Plasmodium falciparum	Plasmodium Vivax	Total
10-25 years	10 (41.66%)	14 (58.33%)	24 (64.86%)
26-50 years	01(14.28%)	06 (85.71%)	07 (18.91%)
>50 years	02 (33.33%)	04 (66.66%)	06 (16.21%)
Total	**13 (35.13%)**	**24 (64.86%)**	**37(100%)**

Table 3: Sex-wise distribution of positive malaria cases

Sex	Plasmodium falciparum	Plasmodium Vivax	Total
Male	12 (36.36%)	21(63.63%)	33 (89.18%)
Female	01(25%)	03 (75%)	4 (10.81%)
Total	**13 (35.13%)**	**24 (64.86%)**	**37 (100%)**

Table 4: Comparison of Leishman stain (Gold standard) with other methods for the diagnosis of malarialparasites

Method	Leishman Stain	Field's Stain	Antigen detection Assay
Negative	487	493	487
P.falciparum	13	10	13
P.Vivax	24	21	24
Total	**524**	**524**	**524**

Table 5: Comparison of the sensitivity and specificity of the various methods detection with Leishman stain

	Leishman Stain (Gold Standard)	Fields stain	Antigen detection assay
Sensitivity	100%	86.05%	100%
Specificity	100%	98.78%	100%
Positive Predictive value	100%	86.05%	100%
Negative Predictive value	100%	98.78%	100%

DISCUSSION

In present study out of 524 patients, 295(56.29%) were male and 229 (43.70%) were female patients. Out of 524, a total of 37 (7.06%) blood samples were positive for malarial parasite. This is less as compared to the study conducted by Takpere *et al* [11] which showed 13.86% percent positivity, while study conducted by Hymavathi *et al*[12] showed similar incidence i.e.8% in urban population. Similar studies conducted by Pinto *et al*[13] and Nandawani *et al*[14] showed malarial positivity of 10.5% and 15% respectively. Out of these 37 positive samples, 24(64.86%) were positive for plasmodium vivax and 13 (35.13%) were positive for plasmodium falciparum. (Table: 1). No mixed infection was seen during this study period. Study conducted by Hymavathi*et al*[12] relatively shows lower incidence of plasmodium vivax (39.4%) than plasmodium falciparum (52.6%). This could be due to regional differences within the tropical areas. Maximum infection was detected in the age group of 10-25 years (64.86%),followed by 26-50 years (18.91%) and more than 50 years (16.21%) (Table:2). Of 37 positive samples, maximum positivity was seen in male (33/37) accounting for 89.18% and 10.81%in females (4/37).This sex distribution showed that males are more prone to the infection which is in accordance with the Nwaorgu and Orajiaka (2011) and Okafor and Oko-Ose, (2012) who reported higher prevalence in males in different parts of the country.[15,16] Among males, 63.63%were positive for plasmodium vivax while 36.36% were positive for plasmodium falciparum, while in female 75% were positive for plasmodium vivax and 25% for plasmodium falciparum respectively. (Table: 3). The present study was designed to compare the sensitivity, specificity, positive predictive value and negative predictive values of rapid antigen detection test and field's stain by using Leishman stained PS as a gold standard. It was found that field's stain showed 86.05% sensitivity,98.78% specificity,86.05% positive predictive value and 98.78%% negative predictive value, while rapid test showed 100% sensitivity, specificity, positive predictive value and negative predictive value (Table:4 and Table:5). This is in concurrence with the earlier studies of Azikiwe CCA *et al* and Aubouy A *et al* which showed

that rapid antigen detection test is as specific as PS and appears even more sensitive than PS[17,18].

CONCLUSION

Rapid diagnostic tests showed good performance as a screening test. Therefore it can be recommended for wide-scale usage when microscopy is not available and immediate clinical diagnosis is required.

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