A Comparative Study of Peripheral Blood Smears and Rapid Diagnostic Tests in the Diagnosis of Malaria at a Tertiary Health Care Hospital in Agra.

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Abstract: The aim of the study was to compare the effectiveness of Rapid Diagnostic Test (RDT) and microscopy, in detection of malaria parasite, among suspected patients of malaria from S.N. Medical College, Agra. During February to July 2018, a total of 700 blood samples were collected for Peripheral blood smear & Rapid Card Test based on Immuno-chromatography. Peripheral blood smear method showed 112 cases positive for malarial parasite. Of these, 108 cases were positive for P.vivax and 4 cases for P.falciparum. Rapid card test method showed 107 positive for malarial parasite. Of these, 104 cases were positive for P.vivax and 3 cases for P.falciparum. Rapid Card Test failed to detect four cases of P.vivax and one case of P.falciparum which were positive by blood smears. Peripheral blood smear method is considered the gold standard in diagnosis but it is labour intensive and time consuming, therefore, may cause delay in diagnosis. Rapid Diagnostic Card test is easy to use, reliable, simple to interpret and an effective tool for rapid diagnosis of malaria with some limitations like high cost and inability of quantification of parasite. It can also be used as an epidemiological tool for rapid screening of malaria or where the facilities for microscopy are not available.

Keywords: Peripheral blood smear, Rapid Card test, P.vivax, P.falciparum

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I. Introduction

Malaria is caused by protozoan parasites of the genus *Plasmodium*. The most serious type of malaria is caused by *Plasmodium falciparum*, which can also be fatal sometimes. The other human malaria species, *P. vivax*, *P. malariae*, *P. ovale*, and sometimes *P. knowlesi* can cause acute, severe illness but mortality rates are low [1]. Malaria is a significant cause of morbidity and mortality in endemic regions, creating enormous social and economic burdens, whereas it is relatively uncommon in developed countries, where it occurs mainly in travellers returning from endemic regions [2].

Malaria is a widely prevalent parasitic disease affecting 500 million people every year and associated with 2-5 million deaths [3]. The accurate diagnosis of malaria infection is important to reduce severe complications and mortality as malaria cannot be diagnosed clinically because the presenting clinical symptoms and signs mimic other tropical infections and hence, must be confirmed by laboratory diagnosis [4]. Although microscopic detection of parasites on blood smears, has been used as the reference standard for diagnosis of malaria in most laboratories for more than a century, it is an imperfect standard as it is highly dependent on the technical expertise of the microscopist [5]. A key feature of the World Health Organization global malaria control strategy is to rapidly diagnose malaria at district and village level, so that effective treatment can be administered quickly to reduce morbidity and mortality [6]. Rapid diagnostic tests (RDTs) could be considered for diagnosis of malaria in most patients of endemic regions, especially in poor power settings where there is shortage of qualified manpower [7].

II. Materials And Methods

Patients attending the hospital, with symptoms and signs suggestive of malaria formed the study group. A total of 700 patients were included in the study. Blood samples collected from the patients were subjected to thick and thin smear (traditional microscopy), and Rapid Card Test (RECKON DIAGNOSTICS P.LTD-IMMUNOPAK) based on immunochromatography.

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III. Blood Smear Staining Method

LEISHMAN METHOD:

Thick and thin smears were prepared and stained with Leishman's stain and examined for malarial parasites by light microscopy.

2.RAPID DIAGNOSTIC TEST(RECKONDIAGNOSTICS P.LTD-IMMUNOPAK):

Malaria *P.falciparum* and *P.vivax* Antigen test is a rapid, qualitative, chromatographic immunoassay for the detection of *P. falciparum* specific histidine rich protein- II (Pf, HRP-II) and *P.vivax* specific pLDH in human blood sample. This test can also be used for specific detection and differentiation of *P. falciparum* malaria and *P. vivax* malaria in areas with high rates of mixed infections. The test was performed as per the manufacturer's instructions and results were read at the end of 20 minutes.

IV. Results

During the course of six months (February to July 2018), a total of 700 blood samples were taken from suspected patients of malaria for Peripheral blood smear & Rapid Card Test based on Immunochromatography. Among these patients, there were 378 females and 322 males. Maximum samples were from the month of July with positivityrate of 18.49%. Positivity rates of remaining months were February (10/72), March(13/91) April (14/88), May (17/106), & June (26/170).

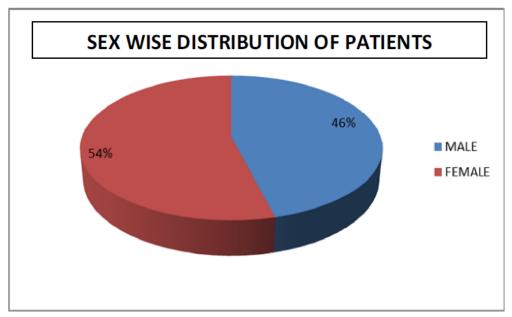


Figure 1- Sex wise distribution of patients

Table 1-Month wise distribution of samples & their result

Months	Blood collection	Smear positive	Smear negative
February	72	10	62
March	91	13	78
April	88	14	74
May	106	17	89
June	170	26	144
July	173	32	141
Total	700	112	588

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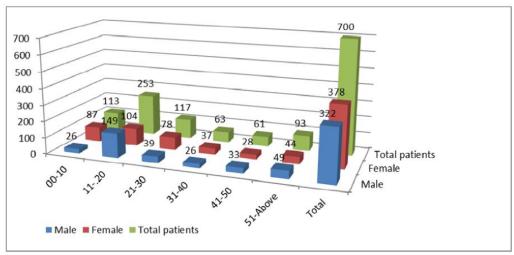


Figure 2- Age Group Distribution of patients

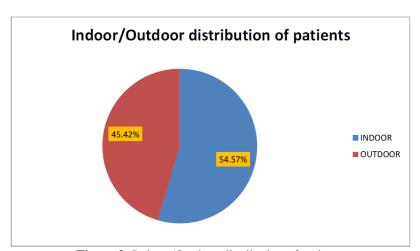


Figure 3- Indoor /Outdoor distribution of patients

Out of 700 samples examined, positivity rate was 16% by peripheral blood smear method, while positivity rate by Rapid card test method was 15.28%. By peripheral blood smear method, 112 cases were found to be positive for malarial parasite. Of these, 108 cases were positive for *P.vivax* and 4 cases were positive for *P.falciparum*. By Rapid card test method, 107 cases were found to be positive for malarial parasite. Of these, 104 cases were positive for *P.vivax* and 3 cases were positive for *P.falciparum*.

Table 2-Comparison of peripheral blood smear with rapid card test method for the detection of malaria parasites

BLOOD SMEAR	RAPID CARD TEST	
P.vivax	+	-
Positive-108	104	4
Negative-592	0	592
Total-700	104	596
P.falciparum		
Positive-4	3	1
Negative-696	0	696
Total-700	3	697

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Table 3-Statistical evaluation of Rapid card test method for the detection of malarial parasites in comparison with Peripheral blood smear method

	RAPID CARD TEST	
	P. vivax	P.falciparum
Sensitivity(%)	100	100
Specificity(%)	99.32	99.85
Positive predictive value(%)	96.29	75
Negative predictive value(%)	100	100

V. Discussion

The present study was done to compare the Rapid card test based on immunochromatography (ICT), with peripheral blood smear examination for the diagnosis of malaria. The peripheral blood smear examination showed that 112 (16%) cases were infected with malaria and 588 (84%) cases were negative. This is comparable with the studies of Dutta G et al [8] and Paul SN et al [9] who reported 16.83% and 16.84% positive cases for malaria parasites respectively, by peripheral blood smear examination. Similarly Sarita Yadav et al [10] in their study reported a positivity rate of 15% for malarial parasites by peripheral blood smear method. Out of these 112 positive cases by peripheral blood smear method, 108 (96.42%) were positive for *P.vivax* and 4 (3.57%) for *P.falciparum*, which is in concordance with the study of Sarita Yadav et al [10] who showed that out of 150 positive cases, 144 (96%) were positive for *P.vivax* and 6 (4%) were *P.falciparum*. Rapid Card Test detected 107 positive cases of malaria out of 700 samples collected from patients suspected of malaria, and 593 cases were negative. Of these 107 positive cases of malaria, 104 were positive for *P.vivax* and 3 were positive for *P.falciparum*. Rapid Card Test failed to detect four cases of *P.vivax* and one case of *P.falciparum* which were positive by blood smears.

Considering Peripheral blood smear method as gold standard for this study, parameters like sensitivity, specificity, Positive Predictive Value (PPV) and Negative Predictive Value (NPV) were calculated. The sensitivity of Rapid Card Test for *P.vivax* was 100% and specificity, Positive Predictive value (PPV), Negative Predictive Value (NPV) were 99.32%, 96.29% and 100% respectively. For *P.falciparum*, the Rapid Card Test showed 100% sensitivity and specificity, Positive Predictive Value (PPV), Negative Predictive Value (NPV) were 99.85%, 75% and 100% respectively. This is in concordance with the study of Sreekanth et al [11] who reported that sensitivity was 100% and specificity, Positive Predictive Value, Negative Predictive Value were 95.7%, 96.3%, 100% respectively for *P.vivax*, while for *P.falciparum* sensitivity was 100% and specificity, Positive Predictive value, Negative Predictive Value were 98.7%, 94.7%, 100% respectively. Dutta G et al [8] in their study also reported a sensitivity and specificity of 100% and 99.70% respectively, by Rapid card test which is comparable with the present study. In the present study, the Positive Predictive Value for *P.falciparum* was lower i.e 75%, which may be due to difference in sample size.

Although no single test can replace the conventional method of peripheral blood smear examination, this newer Rapid diagnosticCard Test can be used as a supplement to microscopic examination of peripheral blood smear where the diagnosis cannot be made on microscopy and an experienced microscopist is not available. The Rapid Card Test is a valuable adjuvant at the time of emergency for rapid diagnosis, whereas microscopy remains the main stay for the diagnosis of malaria.

VI. Conclusion

Malaria continues to be one of the major health problems in the developing world. Accurate diagnosis of malaria is necessary to prevent morbidity and mortality while avoiding the unnecessary use of antimalarial agents. Peripheral blood smear method is considered the gold standard in diagnosis but it is labor intensive and time consuming, therefore, may cause delay in diagnosis. Rapid Diagnostic Card test is easy to use, reliable, simple to interpret and an effective tool for rapid diagnosis of malaria with some limitations like high cost and inability of quantification of parasite. It can also be used as an epidemiological tool for rapid screening of malaria or where the facilities for microscopy are not available. However, Peripheral blood smear method still

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remains superior for accurate species differentiation, quantitation of parasite and maintaining a permanent record.

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